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AN EVALUATION OF THE OYSTER MUSHROOM (*PLEUROTUS FLORIDA*) FOR FOOD PROCESSING POTENTIAL

by

© LORI L. ODDSON

A THESIS

SUBMITTED TO THE FACULTY OF GRADUATE STUDIES AND RESEARCH

IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE DEGREE

OF MASTER OF SCIENCE

DEPARTMENT OF FOOD SCIENCE

EDMONTON, ALBERTA

SPRING 1979

THE UNIVERSITY OF ALBERTA

FACULTY OF GRADUATE STUDIES AND RESEARCH

The undersigned certify that they have read, and recommend to the Faculty of Graduate Studies and Research, for acceptance, a thesis entitled: AN EVALUATION OF THE OYSTER MUSHROOM (PLEUROTUS FLORIDA) FOR FOOD PROCESSING POTENTIAL submitted by LORI L. ODDSON in partial fulfilment of the requirements for the degree of MASTER OF SCIENCE.

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Supervisor

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Date March 27/79

ABSTRACT

In this study the potential of the Oyster Mushroom (Pleurotus florida) for freezing, canning and drying preservation has been explored. The general suitability of the mushroom for industrial processing appears to be due to its ease of cultivation on wheat straw and to low weight and solids losses in blanching.

Freezing studies demonstrated that off-flavor development was extensive in Pleurotus florida frozen in an unblanched state after as little as two months storage. Changes in the lipid components were implicated as the primary cause. Blanching thus appeared to be a necessary pretreatment for freezing. Loss of flavor and a decrease in tenderness were, however, detected in sensory analysis of mushrooms blanched before freezing.

Canning of the stems of Pleurotus florida in a 2% salt brine improved their texture when compared to the fresh stems. Brine canning, on the other hand, was not as suitable for the caps. The use of either cream or butter sauces as canning mediums for the caps resulted in better flavor and overall acceptability scores. Canned cream of mushroom soup formulated with Pleurotus florida compared favorably with a product prepared from the commercially used Button Mushroom (Agaricus bisporus).

Pleurotus florida has been demonstrated to have a low activity of the browning enzyme, polyphenoloxidase. This observation had important implications for drying since

enzymatic discoloration usually presents a major problem in this case. Sun or hot air drying of unblanched Pleurotus florida both produced products with satisfactory characteristics. The flavor of an unblanched sun dried product after storage for ten months at room temperature was found to be indistinguishable from a freshly prepared one; rehydration ratio and color were also excellent. Sun or hot air drying appeared to be among the most promising of the preservation techniques investigated.

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Table of Contents

Chapter — Page

1. INTRODUCTION....1
   1.1 MUSHROOM SUPPLY AND DEMAND....1
   1.2 A CASE FOR THE OYSTER MUSHROOM....2
   1.3 RESEARCH OBJECTIVES....3
2. LITERATURE REVIEW....5
   2.1 THE OYSTER MUSHROOM....5
      2.1.1 Characteristics....5
      2.1.2 Growth Requirements....6
      2.1.3 History and Modern Cultivation....11
         2.1.3.1 Spawn Production....12
         2.1.3.2 Substratum Preparation....13
         2.1.3.3 Growth Phase....14
         2.1.3.4 Large Scale Production....15
         2.1.3.5 Yield and Storage Stability....15
   2.2 CULTIVATION OF OTHER MUSHROOM SPECIES....17
      2.2.1 The Button Mushroom....17
      2.2.2 The Padi Straw Mushroom....20
      2.2.3 Shiitake....22
   2.3 FOOD VALUE OF MUSHROOMS....23
   2.4 FLAVOR CHARACTERISTICS OF MUSHROOMS....28
   2.5 MUSHROOM PROCESSING: INDUSTRIAL PRACTICES AND RESEARCH FINDINGS....30
      2.5.1 Thermal Preservation....30
      2.5.2 Freezing Preservation....33
      2.5.3 Dehydration....37

2.5.4 Novel Processing Methods.....42

2.5.5 Research on the Oyster Mushroom.....42

3. EXPERIMENTAL MATERIALS AND TASTE PANEL SELECTION METHODS.....45

3.1 INTRODUCTION.....45

3.2 CULTIVATION METHOD FOR PLEUROTUS FLORIDA.....46

3.2.1 Preparation of Spawn.....46

3.2.2 Substrate Preparation.....47

3.2.3 Mycelial Phase.....47

3.2.4 Fructification Phase.....47

3.3 YIELD DATA.....56

3.3.1 Methodology.....56

3.3.2 Results.....56

3.4 PROXIMATE ANALYSES.....58

3.4.1 Methodology.....60

3.4.2 Results.....62

3.5 TASTE PANEL SCREENING.....65

3.5.1 Selection of Panelists.....66

3.5.2 Ability to Distinguish Different Mushroom Flavors.....67

3.5.3 Ability to Distinguish Mushroom Flavor Intensity.....67

3.5.4 Ability to Distinguish Textural Differences.....68

3.5.5 Choice of the Final Taste Panel.....68

4. BLANCHING STUDY.....70

4.1 INTRODUCTION.....70

4.2 MATERIALS AND TESTING METHODS.....71

4.2.1 Peroxidase Test.....71

4.2.2 Blanching Tests.....71

4.2.2.1 Blanching Procedures.....72

4.2.2.2 Solids Mass Balance Experiment...73

4.2.2.3 Measurement of Volume Shrinkage..73

4.3 RESULTS AND DISCUSSION.....74

4.3.1 Blanching Time.....74

4.3.2 Water and Steam Blanching Tests.....74

4.3.3 Effect of Post Harvest Storage.....77

4.3.4 Effect of Water Blanching on Solids and Weight Losses.....79

4.3.5 Effect of Blanching on Volume Shrinkage...79

4.4 REMARKS.....82

5. FREEZING EXPERIMENTS.....84

5.1 INTRODUCTION.....84

5.2 COMPARISON OF POLYPHENOLOXIDASE ACTIVITY AND SUBSTRATE LEVELS IN PLEUROTUS FLORIDA AND AGARICUS BISPORUS.....84

5.2.1 Methodology.....85

5.2.2 Results and Discussion.....86

5.3 EFFECT OF FREEZING ON DRIP LOSS, FLAVOR AND COLOR OF BLANCHED AND UNBLANCHED PLEUROTUS FLORIDA.....89

5.3.1 Methodology.....90

5.3.1.1 Measurement of Drip Loss After Thawing.....90

5.3.1.2 Color Evaluation.....90

5.3.1.3 Flavor Evaluation.....91

5.3.2 Results and Discussion.....91

5.4 THE ROLE OF LIPIDS IN THE DEVELOPMENT OF OFF-FLAVOR DURING FROZEN STORAGE OF UNBLANCHED PLEUROTUS FLORIDA.....95

5.4.1 Relationship of Peroxide Numbers and Sensory Evaluation Scores.....95

5.4.1.1 Methodology.....95

5.4.1.2 Results and Discussion.....97

5.4.2 Effect of Frozen Storage on Chemical Changes to the Lipid.....97

5.4.2.1 Methodology.....97

5.4.2.2 Results and Discussion.....100

5.5 TASTE PANEL EVALUATIONS OF FRESHLY FROZEN BLANCHED AND UNBLANCHED PLEUROTUS FLORIDA CAPS..105

5.5.1 Methodology.....105

5.5.2 Results and Discussion.....106

5.6 REMARKS.....108

6. CANNING EXPERIMENTS.....109

6.1 INTRODUCTION.....109

6.2 METHODOLOGY.....109

6.3 RESULTS AND DISCUSSION.....112

6.3.1 Sensory Analysis of Fresh Caps and Stems of Pleurotus florida.....112

6.3.2 Sensory Analysis of Brine Canned Caps and Stems of Pleurotus florida.....113

6.3.3 Sensory Analysis of Cream and Butter Sauce Canned Pleurotus florida.....116

6.3.4 Cream of Mushroom Soups: Comparison of Pleurotus florida and Agaricus bisporus..117

6.3.5 Determination of Weight Loss After Brine Canning: Pleurotus florida versus Agaricus bisporus.....119

6.4 REMARKS.....119

7. DRYING EXPERIMENTS.....122

7.1 INTRODUCTION.....122

7.2 DRYING METHODS AND PRODUCT CHARACTERISTICS.....123

7.2.1 Sun Drying.....123

7.2.2 Hot Air Drying.....123

7.2.3 Freeze Drying.....124

7.2.4 Other Drying Methods.....124

7.3 TASTE PANEL EVALUATIONS OF FLAVOR AND TEXTURE OF FREEZE DRIED AND SUN DRIED PLEUROTUS FLORIDA.125

7.3.1 Methodology.....125

7.3.2 Results and Discussion.....126

7.4 EFFECT OF STORAGE ON SUN DRIED PLEUROTUS FLORIDA.....128

7.4.1 Methodology.....128

7.4.2 Results and Discussion.....129

7.5 REMARKS.....131

8. CONCLUSIONS AND RECOMMENDATIONS.....133

8.1 SUMMARY OF RESEARCH FINDINGS.....133

8.2 SUGGESTIONS FOR FURTHER WORK.....136

9. REFERENCES.....139

10. APPENDIX A.....153

11. APPENDIX B.....161

LIST OF TABLES

Table		Page
1	Shelf life of *Pleurotus ostreatus* and *Pleurotus florida* at 2° and 17°C	16
2	Comparison of the vitamin and mineral content of fresh mushrooms (*Agaricus bisporus*) and some common raw vegetables	26
3	Comparison of the proximate composition of *Agaricus bisporus* and *Pleurotus* species (fresh basis)	27
4	Yield data for first, second and third flush crops of *Pleurotus florida* (fresh weight)	57
5	Weight proportion of caps to stems in *Pleurotus florida* fruit bodies (first flush crop)	59
6	Proximate composition of *Pleurotus florida* caps and stems from first and second flushes	63
7	Test for peroxidase activity in *Pleurotus florida* after steam blanching treatments	75
8	Steam and water blanching of *Pleurotus florida* and *Agaricus bisporus*: effect on % T.S. and weight loss (mean ± standard error)	76
9	Solids and total weight losses of *Pleurotus florida* and *Agaricus bisporus* after water blanching	80
10	Volume shrinkage of *Pleurotus florida* and *Agaricus bisporus* as effected by 'blanching'	81
11	Lightness of blanched and unblanched *Pleurotus florida* after four months frozen storage at -30°C	94
12	Mean taste panel scores for flavor of blanched and unblanched *Pleurotus florida* after four months frozen storage at -30°C	94

Table | Page

13 Summary of results (mean scores) from the sensory evaluation of freshly frozen blanched and unblanched Pleurotus florida as compared to a fresh reference.....107

14 Summary of results (mean scores) from the sensory evaluation of fresh caps and fresh stems of Pleurotus florida.....114

15 Summary of results (mean scores) from the sensory evaluation of canned caps and canned stems of Pleurotus florida.....114

16 Summary of results (mean scores) from the sensory evaluation of fresh and canned caps of Pleurotus florida.....115

17 Summary of results (mean scores) from the sensory evaluation of fresh and canned stems of Pleurotus florida.....115

18 Summary of results (mean scores) from the sensory evaluation of brine, butter sauce and cream sauce canned Pleurotus florida caps.....118

19 Summary of results (mean scores) from the sensory evaluation of soups made from Pleurotus florida caps, Pleurotus florida stems and Agaricus bisporus.....118

20 Summary of results (mean scores) from the sensory evaluation of sun dried Pleurotus florida as compared to a fresh reference.....127

21 Rehydration ratio, time and color of blanched and unblanched sun dried Pleurotus florida after storage.....130

LIST OF FIGURES

Figure		Page
1	Relative activity of the enzyme polyphenoloxidase in *Pleurotus florida* and *Agaricus bisporus* as measured by the extent of browning	87
2	Relative activity of phenolic substrates in *Pleurotus florida* and *Agaricus bisporus* as measured by the rate of browning	88
3	Effect of blanching and freezing rate on drip loss of *Pleurotus florida* after thawing	92
4	Relationship of peroxide numbers and mean taste panel flavor scores for unblanched frozen *Pleurotus florida*	98
5	Comparison of saturated and unsaturated fatty acid methyl esters (FAMEs) from the neutral lipid fraction of fresh *Pleurotus florida* and frozen unblanched *Pleurotus florida* stored for 2 months at -30°C	102
6	Comparison of saturated and unsaturated fatty acid methyl esters (FAMEs) from the phospholipid fraction of fresh *Pleurotus florida* and frozen unblanched *Pleurotus florida* stored for 2 months at -30°C	103
7	Weight loss after blanching and brine canning of *Pleurotus florida* and *Agaricus bisporus*	120

LIST OF PLATES

Plate		Page
1	*Pleurotus florida* spawn	49
2	Apparatus for chopping straw substrate	50
3	Appearance of substrate before and after chopping	50
4	Substrate soaking operation	51
5	Dark chamber for mycelial growth phase	51
6	Inoculated substrate during the mycelial growth phase	52
7	Light chamber for fructification phase	53
8	*Pleurotus florida* fruit bodies (top view)	54
9	*Pleurotus florida* fruit bodies (side view)	55
10	TLC chromatogram of lipid extracts from fresh and stored frozen unblanched *Pleurotus florida* fruit bodies	104

1. INTRODUCTION

Although it is difficult to speculate just how long mushrooms have been consumed as a human food, it is believed to be a practice dating back to Roman times. Hundreds of species are known to be edible, but of these only Agaricus bisporus (The Button or Common Cultivated Mushroom), Vovariella volvacea (The Padi Straw Mushroom), Lentinus edodes (Shiitake), and Auricularia polytricha (The Chinese Ear Fungus) are commercially important (Ooraikul and Mei, 1977). The first three species are cultivated; the latter can be cultivated by primitive methods but is usually collected in the wild (Gray, 1970). The fact that so few mushroom species have been cultivated to date is largely due to problems in replicating the environmental conditions needed. However, it is also true that the above species are very popular; Agaricus bisporus is by far the most widely accepted.

1.1 MUSHROOM SUPPLY AND DEMAND

In recent years, mushroom production and consumption have risen appreciably. So much so, that mushrooms can no longer be considered the luxury item they once were.

Between the years 1950 and 1963 world production and consumption of Agaricus bisporus increased by approximately 300% (Longhill, 1969). In Canada, the market for Agaricus bisporus has been estimated to have increased by over 50%

for the period 1964 to 1969 (Burns and Curry, 1971) and a further 100% between the years of 1969 to 1974 (Agriculture Canada, 1975a). Canadian per capita consumption rose from 1 kilogram in 1969 to 2 kilograms in 1973 (Agriculture Canada, 1975b). It is interesting to note that although mushrooms are Canada's third largest horticultural crop, demand exceeds supply (Burns and Curry, 1971). This is primarily due to the high capital costs associated with the production of Agaricus bisporus and the labor intensiveness of the business. Consequently, about 15% of the total fresh supply is imported from the United States (Agriculture Canada, 1975a). In addition, large amounts of canned mushrooms are also consumed; over 60% of these are imported. Most of the canned imports come from countries like Taiwan and South Korea (Hayes, 1976). Over 99% of the mushrooms grown in the Far East are canned; almost all of these are Agaricus bisporus destined for the export market (Kinrus, 1976).

1.2 A CASE FOR THE OYSTER MUSHROOM

In view of the brisk demand and relatively short supply of Agaricus bisporus, a case could be made for the introduction of other varieties of mushrooms for production in this country. The Oyster Mushroom, now being commercially produced in some European and Asian countries, is suggested for a number of reasons. In the first place, cultivation of this mushroom is easier and more economical than methods presently in use for Agaricus bisporus (Stanek, 1974). This

is primarily because growth substrates (eg. grain straw) do not have to be composted or supplemented with nutrients thus reducing both process steps and costs. Secondly, once the fruit bodies have been removed, the spent substrate is left with an increased protein content and may be a viable material for animal feed (Zadrazil, 1977).

Large volumes of wheat straw are generated annually in North America as a byproduct of agricultural grain production. The Oyster Mushroom has the ability to break down celluloytic material (Toyama and Ogawa, 1976) and is also believed to be capable of the fixation of atmospheric nitrogen (Ginterova, 1973; Ginterova and Maxianova, 1975). The prospect for the application of this fungus in the utilization of straw is probably its most significant characteristic from a cultivation standpoint.

1.3 RESEARCH OBJECTIVES

Much of the research on the cultivation of the Oyster Mushroom, Pleurotus spp., orginated in Europe. In North America, almost no attention appears to have been devoted to this mushroom even though its adaptation to wheat straw cultivation should make it attractive.

Growth requirements and cultivation methods for the Oyster Mushroom have been described in the literature fairly thoroughly, but little is known about the degree of the acceptability of this mushroom as a fresh or processed product. Of particular interest is its adaptability to the

conventional processing methods: canning, freezing and drying. To date, very little research has been specifically concerned with these aspects. With an upsurge of interest in the species and the possibility that it might eventually be produced on a more widespread basis, the need to examine its processing potential is apparent.

The present work has explored the freezing, canning and to a lesser extent drying preservation potential of a variety of the Oyster Mushroom known as Pleurotus florida. Specifically, the objective of the work was to measure certain physical, chemical and sensory changes introduced by the processing methods under examination. The parameters of particular interest were flavor, color and texture. Both subjective and objective tests were used in the overall evaluation.

It should be noted that the scope of this project placed certain constraints on the detail with which each aspect was examined. The general broad approach of these investigations, however, was by design and is hoped not to have limited the usefulness of the work as a whole.

2. LITERATURE REVIEW

2.1 THE OYSTER MUSHROOM

There are several species of the genus *Pleurotus* among which are included: *Pleurotus ostreatus*, *Pleurotus florida*, *Pleurotus cornucopiae*, *Pleurotus eryngii*, *Pleurotus pulmonarius*, *Pleurotus sapidus*, and *Pleurotus sajor-caju*. *Pleurotus ostreatus*, however, appears to be the best known and most widely studied.

2.1.1 Characteristics

Pleurotus ostreatus (Fr.) Kummer, of the Basidomycetes classification, belongs to the Agaricaceae family, genus *Pleurotus*. It has an irregular form, with the stem (stipe) usually placed at one side of the 'umbrella-like' cap (pileus). Because of its similarity in shape to that of an oyster, it has come to be known commonly as the Oyster Mushroom. (Zadrazil, 1976)

Pleurotus ostreatus has a moist, white flesh with agreeable odor and flavor when not fully mature. Its surface color is ash-like grey to brown. Size of the individual mushrooms (also called fruit bodies or sporophores) may vary but on the average ranges from about 7 - 15 cm in cap diameter. The lamellae are broad, decurrent and are whitish or grey in color. Stems are usually very short and positioned either latterally or eccentrically. The species grows in many natural habitats around the world, usually in

overlapping clusters on dead or decaying deciduous trees. Spore deposits are white to lilac-grey in color. (Groves, 1962)

The Pleurotus species which grows in Florida, sometimes also referred to as Pleurotus ostreatus white-type, differs from Pleurotus ostreatus grey-type because of its temperature requirement for fruit body induction. In addition to color, it also has certain other physiological differences such as smaller caps and lower dry matter content (Kalberer, 1976). At the present time, there is some debate over the species rank and interspecific taxonomy of this mushroom. It was first described by Block et al. (1959) and Eger (1965) and is often referred to in the literature as either Pleurotus ostreatus var. Florida or Pleurotus florida. According to Zadrazil (1976) it may be related or identical to Pleurotus cornucopiae. On the other hand, Bresinsky et al. (1976) have stated: "The fact that this strain 'Florida' interbreeds with Pleurotus pulmonarius...shows that the former cannot be taken as an independent species." In this work, the name Pleurotus florida has been used.

2.1.2 Growth Requirements

A number of researchers have investigated the basal nutritional requirements of Pleurotus spp. mycelia on synthetic media (Koch, 1958, Eger, 1970 Voltz, 1972; Cailleux et al., 1976; Macaya-Lizano, 1975; Kostadinov

et al., 1972; Hashimoto and Takahashi, 1976). Various carbon sources (i.e. starch, monosaccharides, disaccharides, ethanol, organic acids etc.) and nitrogen sources (i.e. ammonium and nitrate compounds, amino acids, peptone, urea etc.) have been found to support growth in differing degrees.

Although Voltz (1972) found no difference in mycelial growth in an experiment where 10 vitamins and 5 growth promoting substances were tested against a control, Michalik et al. (1974) reported that small amounts of certain vitamins did improve mycelial growth. Some workers have also reported specific mineral requirements (Hashimoto and Takahashi, 1976; Jandiak, 1976).

Cultivation of Pleurotus spp. can be carried out on a number of materials, eg. grain straw, sawdust, pulp and paper wastes, corncobs and various other food and industrial wastes. In practice, however, straw substratum "... has proved to be the simplest nutrient base." (Zadrazil, 1976)

The addition of various nutrient materials (i.e. organic and/or inorganic nitrogen, mineral salts, carbohydrates, and vitamins) to substrate materials has been investigated by several workers (Block et al., 1958; Block et al., 1959; Bano and Srivastava, 1962; Stanek, 1976; Jandiak and Kapoor, 1976a; Cailleux and Diop, 1976). The conclusion reached by Zadrazil (1976) is that most of these additives do not speed mycelial growth to any appreciable extent, nor do they have a positive effect on yield of fruit

bodies produced from the enriched substrate. Ginterova (1973) and Ginterova and Maxianova (1975) claim that Pleurotus ostreatus has the ability to fix atmospheric nitrogen, thereby accomodating for the rather low levels of nitrogen present in the substrate materials. The work of Rangaswami et al. (1975) supports this conclusion. Furthermore, Pleurotus spp. have the enzymes needed to utilize lignin and cellulose as energy sources (Toyama and Ogawa, 1976).

The substrate pH optimum for the Pleurotus genus as a whole is between 5.0 to 6.5 (Zadrazil, 1976), but is much more narrowly defined for the individual species (Koch, 1958). The addition of substances to the growth media must be considered as to their effect on pH.

The mycelial growth rate of the Pleurotus genus is temperature dependent, with an optimum in the range of 20 to 30°C. However, individual species differ in their temperature requirements for fruit body induction. For instance, Pleurotus ostreatus requires a 'cold shock' treatment; temperatures of 15°C or lower must be provided for fructification (Stanek and Rysava, 1971). Pleurotus florida, on the other hand, will fruit at temperatures of up to 27°C (Kalberer and Kunsch, 1974). In either case, the higher the temperature the lighter will be the color of the sporophores produced (Zadrazil, 1976).

Gaseous environment is also important. High concentrations of carbon dioxide have been shown to have a

stimulatory effect on the growth of Pleurotus spp. mycelium (Zadrazil, 1976). Under these conditions, mycelial growth easily 'outcompetes' any invading microorganisms (Schanel et al., 1974; Zadrazil and Schneidereit, 1972; Zadrazil, 1975). Zadrazil (1976) has demonstrated that a carbon dioxide concentration of up to 28% the volume of the air will stimulate mycelial growth. In fact, it is thought that carbon dioxide concentration is the key to the biotechnology of Pleurotus cultivation (Zadrazil and Schliemann, 1976). It should be pointed out, however, that while mycelial growth prefers a semi-anaerobic environment, the development of fruit bodies will take place only under fully aerobic conditions (Schanel et al., 1974); facilities for good air exchange must be provided at this time.

Light is necessary for primordial initiation and subsequent development of fruit bodies (Kalberer, 1976; Gyurko, 1972; Eger et al., 1976a; Eger et al., 1976b; Zadrazil and Schneidereit, 1972). Gyurko (1972) states that once mycelial development is complete, low intensity light of over 40 lux should be provided. Eger et al. (1976a) stress that regular and rapid development of sporophores occurs only when continuous low intensity light (200 - 400 lux) is applied. Eger et al. (1976b) have also demonstrated that sporophore color is dependant on light levels, in addition to temperature as previously discussed. Accordingly, the higher the light intensity and lower the temperature, the darker is the resultant color of the fruit

body. It has also been shown that pileus expansion depends on adequate light provisions. If light levels are too low the stipe becomes increasingly elongated and pileus development does not proceed normally (Eger et al., 1976b). Kalberer's experiments (1976) showed that in the range of 4 - 430 lux, yields of fruit bodies are fairly constant but that the ratio of stem to cap changes markedly (i.e. the lower the light intensity, the greater is the proportion of stem growth).

During the mycelial growth phase a covering material is normally employed to reduce moisture loss. Hence, there is no rigid requirement for air humidity provisions at this time. However, relative humidities of approximately 80 to 95% must be provided for proper fructification (Zadrazil, 1973a, 1973b). If humidity is either too high or too low over the fructification period, abnormal fruit bodies containing excessive or deficient amounts of water will result.

From the foregoing discussion, it is apparent that the conditions for mycelial growth and fructification of Pleurotus spp. are fairly well defined. In addition, studies to date have provided us with the technology for successful cultivation of this fungus. If air temperature, humidity, light intensity, and carbon dioxide concentration are provided at or near optimal amounts, and a suitable growth medium is used, well formed fruit bodies (with small or almost non-existent stipe and large uniform-shaped pilei)

can be produced with relative ease.

2.1.3 History and Modern Cultivation

In the early part of the twentieth century a method for the cultivation of Pleurotus ostreatus on tree stumps was described by Brasch, Busse, and Falck (Zadrazil, 1976). But, it was not until the work of Block et al. (1958) and Block et al. (1959) that a more suitable means of cultivation was developed. Block et al. (1958) used a sterilized mixture (30 min.; 121°C) of oatmeal-fortified sawdust as the substrate for a Pleurotus species. One gallon cans of the sterilized substrate were inoculated with 21 grams of spawn per can. These were stored at 26°C and 85% R.H. for 10 - 14 days at which time fruit bodies began to appear. Block et al. (1959) reported that of nine species of wood destroying basidomycetes tested, "Pleurotus ostreatus promised the most vigorous growth and ability to fruit readily".

In 1962, Bano and Srivastava tested several substrates: paddy straw, rice hulls, horse-manure, sawdust and various combinations of these, and reported that paddy straw gave the highest yields of Pleurotus. In their process, soaked bundles of paddy straw were thinly layered with spawn bits distributed at the periphery of each layer. The 'beds' were supplemented with oatmeal, pulse powder or inorganic nitrogen and watered twice daily. Oatmeal was found to be the best source of added nutrients on the basis of increased yield. The variety of mushroom used was found to withstand

wide temperature and humidity ranges (21-33°C; 67-72% R.H.). Park et al. (1975) also investigated the cultivation of Pleurotus ostreatus on rice straw. They found that this method resulted in better yields than traditional cultivation methods on tree logs.

Eger (1965, 1970, 1976; Eger et al., 1976a, 1976b) has worked extensively with Pleurotus ostreatus and Pleurotus florida. He is well known for his research on the genetics of the fungus, as well as for his contributions to the understanding of fundamental growth requirements and characteristics.

In 1969 Toth cultivated Pleurotus ostreatus on crushed corn cobs under sterile conditions and shortly thereafter the method was modified by Gyurko so that it could be adapted to non-sterile conditions (Kostadinov et al., 1972). Kaul and Janardhanan (1970) successfully produced Pleurotus fruit bodies on chopped branches of Euphorbia royleana, a tree which grows abundantly in India.

Modern culture techniqes have been described by Kalberer (1976), Kalberer and Vogel (1974), Zadrazil (1973a, 1976), Zadrazil and Schneidereit (1972) Laborde and Delmas (1974), Omori (1976), and Weijer (1974). These works have provided the basis for the discussion which follows.

2.1.3.1 Spawn Production

A number of materials can be used for preparation of Pleurotus spawn. These include: wheat kernels, rye kernels,

millet, finely shredded straw, perlite coated with wheat flour, rice and/or wheat bran (Weijer, 1974). Regardless of which inoculum material is chosen, moisture content must be raised to approximately 70% either by soaking or cooking in water (Jablonsky, 1974). The wetted material is then sterilized (121°C) and cooled before being aseptically inoculated with mycelial culture. Following incubation at 25 to 30°C for 10 - 14 days the spawn is ready for use. Jars of spawn thus prepared can be stored under refrigeration (2 - 5°C) for future use. Dudka et al. (1976) have described a complete procedure for the production and storage of Pleurotus ostreatus mycelium.

2.1.3.2 Substratum Preparation

Straw to be used must first be chopped (approximately 5 - 10 cm lengths or smaller) and soaked to obtain a final moisture content of 65 - 75% (Jablonsky, 1974). Additional substances if they are to be used should be added at this point. The straw or straw mixture is then either pasteurized or sterilized. Stanek (1976) recommends a pasteurization procedure (50-70°C for 1 to 2 days) which permits the proliferation of thermo-tolerant sporoform bacteria (B. subtilis, B. mesentericus, and B. macerans). These have been found to be stimulatory for mycelial growth of the Pleurotus fungus. According to Gyurko (1974), the effect is due to a prohibitive action on mold growth. Other methods of heat treatment which have been proposed are: 121°C for 8 hours, 4

- 5 days at 72°C and 60°C for 50 hours (Zadrazil and Schneidereit, 1972). After cooling, the heat treated substrate is ready for spawning.

2.1.3.3 Growth Phase

Kalberer (1976) and others have suggested the use of approximately 3% (w/w) spawn inoculum (wet weight basis). The inoculum should be uniformly dispersed into the substratum material in a manner not conducive to the introduction of contaminants, particularily molds. Polyethylene bags have been found to be convenient for the inoculation and incubation steps . As discussed, the high carbon dioxide atmosphere formed by this type of regime, will stimulate mycelial growth (Zadrazil and Schneidereit, 1972). The plastic bags containing inoculated substrate are pressed firmly into containers of suitable size and then incubated (20 - 30°C at 60% R.H.) for approximately 2 weeks (Weijer, 1974). By this time, dense mycelial growth will have permeated the straw forming coherent 'blocks' which are then uncovered and placed in a facility where light, fresh air and humidity (80 - 90% R.H.) can be provided (Kalberer and Vogel, 1974). Temperature of this facility should be set at 15°C for Pleurotus ostreatus and 20°C or higher for Pleurotus florida (Zadrazil, 1976). Fructification normally occurs in about 7 - 14 days. Several crops ('flushes' or 'breaks') can be obtained but a dark resting phase (10 days) is required prior to light re-introduction (Weijer, 1974).

2.1.3.4 Large Scale Production

Zadrazil (1976) has developed a method suitable for large scale Pleurotus cultivation which includes a process for continual substratum preparation and an improved container for the substratum. An automated commercial operation in Bratislava, Czechoslovakia is currently supplying local and export markets; it has almost displaced the production of Agaricus bisporus in the immediate vicinity (Weijer, 1978).

2.1.3.5 Yield and Storage Stability

Yields of Pleurotus ostreatus are considered high on the basis of weight of mushrooms harvested per weight of substrate used (Kurtzman, 1975; Block et al., 1958). Block et al. (1958), in small scale yield experiments, found that Pleurotus ostreatus produced about 40% more mushrooms than Agaricus bisporus per dry weight of substrate utilized.

Shelf life studies on Pleurotus ostreatus and Pleurotus florida were conducted by Gormley and O'Riordain (1976). Their experiments measured weight loss, color and texture of the fresh mushrooms after storage at either 2°C or 17°C. Samples were packaged in three ways: 1.) on fiberboard trays (uncovered), 2.) on fiberboard trays (covered with PVC film), 3.) in polyethelyene bags sealed with twist ties. Results for weight loss according to the method of packaging are shown in Table 1.

As might be expected, weight loss was less in a given

Table 1 Shelf life of *Pleurotus ostreatus* and *Pleurotus florida* at 2° and 17°C[1].

*Pleurotus florida** (weight in grams)

	Day	UC	C	P
2°C	0	100	100	100
	2	88	96	99
	4	72	95	99
	6	55	94	99
	8	47	93	99
	10	--	93	99
17°C	0	100	100	100
	1	--	--	--
	2	71	94	99

*Pleurotus ostreatus** (weight in grams)

	Day	UC	C	P
2°C	0	100	100	100
	2	69	95	100
	4	43	94	100
	6	26	91	99
	8	--	90	98
	10	--	88	98
17°C	0	100	100	100
	1	--	--	--
	2	62	92	100

* UC=uncovered; C=covered; P=polyethylene bagged

[1]From Gormley and O'Riordain (1976)

time period at 2°C and covering greatly reduced moisture loss. Color and texture changed little under either regime and flavor was considered excellent after 10 days storage at 2°C. These results suggest that the Oyster Mushroom has favorable storage capabilities as a fresh product.

2.2 CULTIVATION OF OTHER MUSHROOM SPECIES

Without sufficient knowledge of how other mushroom species are cultivated, it is difficult to appreciate the ease with which the Oyster Mushroom can be grown. Thus, in the following section, cultivation methods for the commercially significant species will be briefly reviewed. According to Singer (1961), the three mushrooms of major importance are: Agaricus bisporus (Common Cultivated White or Button Mushroom), Volvariella volvacea (Padi Straw Mushroom), and Lentinus edodes (Japanese Shiitake).

2.2.1 The Button Mushroom

The cultivation of the Button Mushroom (Agaricus bisporus), usually credited to the French, dates back more than 300 years. Since then, many technological advances have been made. The process used today is highly sophisticated and complex in nature; a considerable amount of information on cultivation procedures and techniques is currently available. References for this summary included: Singer (1961), Gray (1970), Smith (1969) and Hayes (1972, 1974, 1976).

The three basic phases of the cultivation cycle are:

a. spawn production,

b. substrate composting,

c. fruit body production.

Pure spawn is prepared by inoculating jars of a sterile substrate (composted horse manure was the original type; grain and tobbacco substrates are the two other widely used types) with a small amount of master culture and allowing the containers to 'run' at about 21°C until thoroughly permeated with mycelium. This having been done, the spawn is ready for use.

Once a supply of good spawn is assured the next step is composting of the substrate on which the mushrooms will be grown. Composted horse manure, the traditional substrate, is still used today for this purpose. Other substances (including gypsum, phosphate, and a nitrogen source), added at different stages of the process, perform various functions. Correct proportions of wetted straw, manure and other ingredients are carefully mixed and stacked into high piles. The piles are turned inward and watered at approximately two to three day intervals. The entire composting process takes about 15 days, the end being signalled when the pH (which may reach a maximum of 9) begins to drop and temperature reaches 70 to 80°C. When the process is complete the color of the compost should be a dull brown, all straw particles should be completely decomposed, and no faecal or ammoniacal smell should be

detectable. At this stage the compost is ready to be packed into wooden trays or boxes and stacked in a sealed room where temperature control is available. When its temperature has again risen to 58 - 60°C (about 3 to 5 days), air temperature in the 'sweating-out' room is carefully regulated so that 'peak heat' is maintained for approximately 12 hours. The entire 'sweating-out' process takes about 7 to 10 days. Compost, pasteurized in this way, provides the necessary nutrients for mushroom growth, and as well, is a selective medium free from pathogenic organisms and other undesirable competitors.

When the temperature of the compost has fallen to about 22°C it is ready for spawning. This is usually done by broadcasting or mechanical mixing. The inoculated trays are incubated in dark humid rooms (95% R.H.) for 2 - 4 weeks at 20 - 24°C. By this time the spawn should be well run through the compost. Air temperature is then reduced to 15°C, relative humidity is set at 90% and the trays are 'cased' with a soil mixture (3 - 4 cm thickness). The casing is the site of the critical change that occurs when the mycelium forms a primordium which subsequently develops into a fruiting body. Pseudomonas putida, a soil organism, has been found to be active in promoting fructification. Plenty of fresh air must be provided since it has been shown that carbon dioxide toxicity arises mainly during this period. After three weeks in the mushroom house the first crop should be ready for picking.

Several additional flushes, each with diminishing yields, will be produced over the total cropping period, but most mushroom growers stop harvesting after the third or fourth flush. The average yield per square meter of mushroom bed is approximately ten kilograms.

According to Gormley (1975), Agaricus bisporus can be stored for 5 days at 1°C followed by 2 days at 20°C. Langerak (1972) states that for maintenance of high quality, Agaricus bisporus should only be stored for 3 days at 3 to 5°C. Sveine et al. (1967) have cautioned against storage temperatures below 3°C. The merits of various packaging methods and materials on the keeping quality of fresh Agaricus bisporus have been discussed by Sveine et al. (1967), Langerak (1972), and Nichols and Hammond (1975).

2.2.2 The Padi Straw Mushroom

Volvariella volvacea, the Padi Straw Mushroom, is cultivated in tropical and semi-tropical climates in China and Southeast Asia (San Antonio and Fordyce, 1972). Although the art of its cultivation is very old, methods used today are much less refined than those for Agaricus bisporus (Singer, 1961; Gray, 1970; San Antonio and Fordyce, 1972). Nevertheless, considerable advances have been made in recent years to improve the techniques.

The traditional procedure for outdoor cultivation consists of the following basic steps:

a. land preparation,

b. rice straw soaking,

c. bed construction and spawning

d. growth of fruit bodies and cropping.

In contrast to the cultivation of Agaricus bisporus, pure culture spawn methods are not widely used. Even though these have been shown to be superior, the use of 'flake spawn' (spawn obtained from spent compost beds) still persists in Padi Straw Mushroom cultivation (Ho, 1972).

The first step to be undertaken is plowing and flooding of the land to be used for the mushroom beds. After draining and partial drying, elevated soil bases (about 0.1 m high, 1.3 m long and 0.35 m wide) are made by hoeing the soil into beds in an east west direction. (Gray, 1970). Next, rice straw is tied into bundles weighing approximately two kilograms each, soaked in water until thoroughly wetted and composted for 3 to 4 days; temperature may reach 64°C (Chang, 1972). These bundles are carefully stacked on the soil beds and inoculated by placing pieces of spawn (10 - 15 cm apart) around the border of each layer as the bed is built up (Singer, 1961). The beds are covered with straw mats or polyethylene sheets and water is added as necessary to keep moisture at the correct level (Gray, 1970). Mushrooms should begin to appear 10 to 12 days later (Ho, 1972). They are usually picked in the 'button' stage before the universal veil opens and this may necessitate two or more pickings per day (Gray, 1970). Chang's experiments (1972), under controlled laboratory conditions, have shown

that the air temperature should be maintained at 28 plus or minus 2°C and relative humidity kept at 85 - 95%; bed temperature should be 34 - 37°C with a pH of 5 to 6.

As with Agaricus bisporus, several successive flushes occur. The harvest intervals are about 5 - 10 days apart and one cycle will normally last from 35 to 56 days (Chang, 1972). According to San Antonio and Fordyce (1972), yields of the Padi Straw Mushroom can be expected to be roughly five times less than those of Agaricus bisporus; their keeping quality also leaves much to be desired.

2.2.3 Shiitake

Lentinus edodes, more commonly known as Shiitake, fruits naturally on trees in the woods of Eastern Asia (Gray, 1970). Like the Padi Straw Mushroom, it has been cultivated for centuries but modern cultivation methods are somewhat more sophisticated. The basic procedure for growing Shiitake is fairly similar to either of the methods for the two mushrooms previously discussed, except that there is no 'bed' preparation per se.

The first phase in the operation is the gathering of wood (eg. oak, chestnut, beech), the medium on which these mushrooms are grown. At the right time of year, the trees must be felled and cut into logs (approximately 1 m long and 5 - 15 cm in diameter) (Singer, 1961). These are then inoculated by one of the following methods: pieces of spawn are inserted into holes drilled in the logs or a spore

emulsion is poured, sprayed or injected into incisions made in the sapwood (Singer, 1961).

The inoculated logs are put into 'laying yards' for 5 to 8 months over which period the mycelium gradually permeates them (Singer, 1961). After this, they are transferred to 'raising yards' where more moisture and a temperature of 12 - 20°C will induce fruiting (Gray, 1970). It should probably be mentioned that not only do 'laying yards' and 'raising yards' differ in the processes performed in them, but the positioning of the logs is also quite different for important reasons.

The 'raising yards' can be expected to produce mushrooms "...for a minimum of three years and may continue production for as long as six years" (Gray, 1970). No care, other than frequent watering and daily harvesting is necessary. Shiitake mushrooms must be picked young otherwise they are not of an acceptable eating quality. No figures on expected yields or storage stability of this mushroom were located in the literature.

2.3 FOOD VALUE OF MUSHROOMS

Mushrooms are frequently regarded as having little or no food value. In fact, this is not the case. It is true that their caloric value is quite low (most mushrooms contain about 90% water) but in terms of nutrient content they fare as well as, if not better than, most plant materials.

Mushrooms are a good source of protein, normally containing from 2 - 4% depending on the species (Kurtzman, 1975). It should be noted, though, that crude protein values tend to overestimate the amount of protein present since chitin, nucleic acids and other nitrogenous compounds contribute to the nitrogen analysis. Kurtzman (1975) has proposed that N X 5.16 would give a more accurate estimation of the protein content. Nevertheless, the protein value of a number of mushroom species is estimated to be approximately double that of some vegetables, eg. asparagus, cabbage and potatoes (Chang, 1972). All of the essential amino acids are present in Agaricus bisporus, although methionine is usually found in relatively small amounts (Weaver et al., 1977). Chromatographic studies of four other mushroom species grown on synthetic medium revealed that the common amino acids are presents in comparable amount (Purkayastha and Chandra, 1976). Protein digestibility values may range from 69 - 83% (Hayes and Haddad, 1976), again depending on the species. In general, the quality of the protein in mushrooms is intermediate between plant and animal proteins.

The carbohydrate content of Agaricus bisporus has been estimated at 2.45 - 4.75%, a large portion of which arises from the mannitol content (Holtz, 1971). Fat content is very low, usually less than 0.5% (Kurtzman, 1975). Agaricus bisporus is a relatively good source of potassium, phosphorus, and iron but is low in calcium; mineral content is normally about 1.1% (Hayes, 1976). Vitamins A, D and E

are absent and vitamin C is low, but concentrations of niacin and riboflavin are high and thiamine level fair (Gormley, 1973). The data in Table 2 give a comparison of the vitamin and mineral contents of Agaricus bisporus and some common raw vegetables.

The Oyster Mushroom is well balanced in terms of amino acid content with slightly higher values for total sulfur-containing amino acids than those found for the Button Mushroom (Jandiak and Kapoor, 1976b). According to Kalberer and Kunsch (1974), "The amino acid composition of Pleurotus ostreatus and Agaricus bisporus sporophores ... are rather similar". Protein content has been reported to be in the range of 2.3% (Lelley, 1974) to 5% (Ola'h, 1975). Other constituents are also comparable in amount (Andreotti et al., 1975; Bano et al., 1963; Kurtzman, 1975). No data on vitamin and mineral content could be located. Table 3 provides a summary comparison of the proximate composition of Pleurotus spp. and Agaricus bisporus.

Table 2 Comparison of the vitamin and mineral content of fresh mushrooms (Agaricus bisporus) and some common raw vegetables.

	Mushroom[1]	Potato[2]	Carrot[2]	Pea[2]	Corn[2]
	(mg of nutrient/100 g of fresh material)				
Calcium	17	7	37	62	3
Potassium	150	53	36	92	111
Iron	2.7	0.6	0.7	0.7	0.7
Sodium	64	3	47	-	trace
Phosphorus	150	407	341	170	280
Vitamin A*	0	trace	1110	680	400
Thiamine	0.1	0.1	0.06	0.28	0.15
Riboflavin	0.5	0.04	0.05	0.12	0.12
Niacin	4.1	1.5	0.6	-	1.7
Vitamin C	2.5	20	8	21	12

* in International units (I.U.)

[1] Hayes, 1976
[2] Watt and Merrill, 1963

Table 3 Comparison of the proximate composition of *Agaricus bisporus* and *Pleurotus* species (fresh basis).

	Water %	Crude ptn. %	Crude fat %	Crude fiber %	CHO %	Ash %
A. bisporus[1]	90.4	2.7	0.3	0.8	-	0.9
A. bisporus[2]	91.0	3.3	0.4	-	2.4	0.9
A. bisporus[3]	89.7	4.9	0.2	0.4	1.1	0.8
P. ostreatus[4]	90.9	2.8	0.2	1.1	-	1.0
Pleurotus spp.[5]	85-90	3-5	0.4	-	6.0	1.0
Pleurotus spp.[6]	90.9	2.9	0.6	1.1	-	1.0

[1] Watt and Merrill, 1963
[2] Hayes and Haddad, 1976
[3] Chang, 1972
[4] Bano *et al.*, 1963
[5] O'lah, 1975
[6] Kurtzman, 1975

2.4 FLAVOR CHARACTERISTICS OF MUSHROOMS

Research devoted to the isolation and identification of mushroom flavor components has permitted the comparison of the flavor components of various species. As well, some information on the changes which result from processing is now available.

Johnson *et al.* (1971) reported that high protein content contributes to mushroom flavor. They claim that the basic amino acids are largely responsible for this effect. The results of Litchfield's work (1967) with *Morchella sp.* support this finding. Glutamic acid is thought to be the amino acid of primary importance (Dijkstra and Wiken, 1976). Purine bases and nucleotides have also been identified as having an important impact on flavor (Cronin and Ward, 1971) as have carbohydrates (Dijkstra and Wiken, 1976).

Insofar as volatile components are concerned, several reports suggest the L form of 1-Octen-3-ol to be the most important volatile flavor compound in *Agaricus bisporus* (Cronin and Ward, 1971; Wasowicz, 1974; Dijkstra and Wiken, 1976; Dijkstra, 1976). 1-Octen-3-ol has also been identified in a number of other mushroom species although it varies quite markedly in concentration (Dijkstra, 1976). Modifications in flavor are thought to be due to differences in the concentration of 1-Octen-3-ol as well as to the presence of other compounds, volatile and non-volatile alike (Dijkstra and Wiken, 1976).

Whereas 1-Octen-3-ol is believed to be the main mushroom volatile, glutamic acid and 5'-guanine monophosphate (5'-GMP), which are normally classed as 'flavor enhancers', have also been shown to contribute substantially to the flavor. The flavor enhancing capacity of these two compounds is well documented although little is known about their mode of action (Teranishi et al., 1971).

Dijkstra (1976) conducted an investigation in which eight fresh, three canned and five dried mushroom species were compared for concentrations of 1-Octen-3-ol, 5'-GMP and glutamic acid. Fresh Pleurotus ostreatus contained substantially more 1-Octen-3-ol and 5'-GMP than fresh Agaricus bisporus. Glutamic acid was dectected in all fresh samples, but varied in concentration from one species to another. Sterilization and drying of the species investigated (Agaricus bisporus, Boletus edulis, Cantharellus cibaricus, Lentinus edodes, Gyromytra esculenta, Marasmius scorodonius and Tricholoma portentosum) resulted in decreases of 1-Octen-3-ol and 5'-GMP; an exception was dried Shiitake where 5'-GMP was strongly retained. Concentration of glutamic acid increased somewhat after sterilization (possibly due to protein breakdown) and decreased on drying (perhaps because of Maillard reactions). Dijkstra (1976) states that these losses may explain the weaker flavor of some canned and dried mushrooms. According to Wasowicz (1974), thermal preservation causes serious changes to the volatile fraction of mushrooms; benzyl

alcohol was one of the main volatile components detected in boiled samples of Agaricus bisporus.

2.5 MUSHROOM PROCESSING: INDUSTRIAL PRACTICES AND RESEARCH FINDINGS

The main aim of this project was to assess the Oyster Mushroom for its food processing potential. Hence, information on the industrial practices and data on the research findings for processing of this and other mushrooms was of paramount concern. However, a review of the literature revealed that almost all of the practical knowledge related to mushroom processing is based on the use of Agaricus bisporus. Consequently, the information reviewed in this section pertains only to the use of the Button Mushroom (Agaricus bisporus) unless another species name is specifically given.

2.5.1 Thermal Preservation

Canning is by far the most widely used preservation technique for mushrooms in North America. In the U.S. approximately 50% of total mushroom production is utilized by canning operations (Anon, 1974). Similarly, Canadians process over half of their domestic supply by this means (Burns and Curry, 1971). In addition, both countries are major importers of canned mushrooms (Butz et al., 1969; Burns and Curry, 1971). The bulk of canned mushrooms on the market today are Button Mushrooms (Agaricus bisporus),

however, the author has observed a few other imported canned varieties (i.e. Padi Straw Mushrooms, Boletus spp., Chantrelles) which are available in specialty food stores.

The basic steps to be followed in a mushroom canning operation have been outlined by Luh and Woodroof (1975) as follows:

a. Sorting and washing
b. Blanching (steam or water at 100°C; 5-7 minutes)
c. Size grading
d. Can filling and exhausting
e. Retorting (18-20 minutes at 121°C for can size A1)
f. Cooling

Typical commercial mushroom canning operations have been described by a number of authors (Lopez, 1975; Anon, 1974; Anon, 1977).

The blanching process is of particular importance in that it removes tissue gases, inactivates the enzyme polyphenoloxidase (PPO) which is responsible for oxidative browning, improves texture for slicing and reduces bacterial counts (Luh and Woodroof, 1975).

Canned mushrooms are a low acid product and must therefore receive a sterilization process adequate to destroy the spores of Clostridium botulinum as well as those of various thermophillic bacteria. This means that retort temperatures of over 100 °C (usually 118-125°C) must be used. For mushrooms canned in brine (can size A1) an Fo value of 8 - 10 is required; in butter sauce (can size up to

A1) an Fo of 6 - 8 is customary (Brennan et al., 1969).

Several styles of canned mushrooms are available. These include: buttons (whole, sliced or quartered) and stems and pieces. In the U.S. 75% of all canned mushrooms are stems and pieces (Anon, 1974). Among the types of packs which may be utilized, brine is the most common, representing about 90% of the total production (Anon, 1974). Other packs (butter or butter sauce, cream sauce, vinegar, oil and wine) are also used, but to a much lesser extent.

Various additives can be used to improve the functional characteristics of canned mushrooms. For example, ascorbic and/or citric acids are permitted in Canada and the United States to perserve color. Monosodium glutamate, which is used for flavor enhancement, is permitted in the United States but not in Canada.

McArdle and Curwen (1962) conducted experiments in which the effect of thermal processing on mushroom constituents was examined. They found that ash content in canned mushrooms was considerably higher than in fresh counterparts (obviously as a result of salt absorption from the brine) and that carbohydrate content was substantially decreased. Little if any protein or fat were found to be lost during canning. Maggioni and Renosto (1970) also studied changes in composition during canning with similar results.

The biggest problem facing mushroom canners is shrinkage during blanching and sterilization. Weight

reductions in the range of 30 to 40% are common (McArdle and Curwen, 1962; Mehlitz and Geerds, 1968) and this represents a potentially large economic loss.

A considerable amount of research has focussed on ways to reduce these losses. For example, Beelman et al. (1973) developed a process called the PSU-3S process in which shrinkage was substantially reduced by soaking fresh mushrooms in water for 20 minutes, storing at 2°C for 18 hours and then soaking a second time for 2 hours prior to processing. This procedure, however, proved difficult for processors to integrate into commercial operations. Subsequently, McArdle et al. (1974) investigated a vacuum soak process followed by storage at 2°C and reported yield increases of up to 15%. In both of these experiments storage alone (24-72 hours at temperatures ranging from 2 - 22°C) was found to increase yield, but increases in water-holding capacity (WHC) were further enhanced by the combination of storage and soaking treatments. A modification of the vacuum-soak method has been patented by Beauvais and Sindall (1974) and is said to be very effective when used in conjuction with high temperature short time sterilization.

2.5.2 Freezing Preservation

Most vegetables have been highly successful when marketed in the frozen state largely because of improved organoleptic characteristics over canned products. The market for frozen mushrooms, however, has not yet reached a

significant proportion on a commercial scale. Mushrooms are particularly delicate with respect to flavor, color and texture. Hence, their preservation by freezing is not without its difficulties.

The prevention of discoloration before, during and after freezing is one of the major problems with Agaricus bisporus because of its high level of PPO. While it is true that blanching will inactivate the enzyme, it has its drawbacks. As previously discussed, one of the serious disadvantages is the subsequent loss in product weight. In addition, blanching causes mushrooms to take on a somewhat greyish and unappealing color, and as well, may reduce flavor and possibly introduce detrimental textural changes (Luh and Woodroof, 1975). Thus, it would be highly desirable if the blanching operation could be avoided in freezing operations.

A number of workers have reported various methods in which additives are used in place of blanching for improving the color of frozen mushrooms (Fang et al., 1971; Fang et al., 1976a; Hanson, 1975; Gormley, 1972; Fujimoto et al., 1972). These findings are summarized below:

a. 0.1-0.5% sodium metabisulfite improved the color of frozen mushrooms to a greater extent than did EDTA, sodium metaphosphate and L-ascorbic acid (Fang et al., 1976a).

b. Color of mushrooms is preserved by treatment with sulfur dioxide gas (9 - 25% by volume) at ambient

temperatures for 10 seconds to 30 minutes prior to freezing (Fang et al., 1971).

c. Lightness is improved by treatment with citric acid (Gormley, 1972).

d. Carbon monoxide gas aeration (170 minutes) was found to alleviate browning problems and not adversely affect flavor (Fujimoto et al., 1972).

e. Mushrooms treated in an aqueous dip of 400 ppm sulfur dioxide gas, sodium chloride, and 0.5% citric acid (or other carboxylic acid) followed by a partial blanch were found to have a superior appearance (Hanson, 1975).

The maintenance of good flavor characteristics is also a fundamental concern in the freezing preservation of mushrooms. While it is true that freezing per se does not cause significant flavor changes, oxidation of chemical components in storage can result in a loss of flavor, or in some cases, development of off-flavors. A primary function of blanching as a pretreatment for freezing is to inactivate the enzymes which catalyze the formation of these undesirable flavor changes. Lipases and/or lipoxidases are believed to be the enzymes involved in the off-flavor development in unblanched or underblanched vegetables which are subsequently frozen (Ciobanu et al., 1976).

In an investigation conducted by Leach (1964), mushrooms frozen in an unblanched condition were evaluated by a sensory panel after 5 months storage. No deleterious

effects on flavor were apparent from the results. If this conclusion is accurate it implies that blanching is an unnecessary pretreatment for freezing of Agaricus bisporus insofar as enzymatic flavor changes are concerned. It is interesting to note that according to the Canadian Government Specifications Board Regulations (Standards for Frozen Vegetables) (1975), mushrooms are the only product for which "additives may be used in place of the blanching process".

It is also highly desirable that the texture of a frozen product after thawing be as close to that of the fresh product as possible. According to Ciobanu et al. (1976), individual quick freezing (IQF) is highly recommended because rapid freezing rates cause lesser amounts of intercellular ice to be formed and thereby result in reduced drip on thawing. The application of IQF techniques substantially improves the texture of the final product by minimizing cellular tissue damage caused by ice crystal growth (Fennema, 1966).

The effect of freezing method on resultant texture in mushrooms has been investigated by several workers. Fang et al. (1976a) reported that IQF freezing is superior to air blast freezing on the basis of drip loss and organoleptic scores. The results of Astrom and Longdahl (1969) support this conclusion. Holdsworth (1967), has also suggested the use of cryogenic freezing (i.e. liquid nitrogen, freon etc.; immersion or spray techniques) for mushrooms as a means of

better preserving the texture. Gormley (1972), on the other hand, found that Freon immersion freezing was less satisfactory than blast freezing in that the former resulted in substantially more drip on thawing. Commerical operations utilizing liquid nitrogen for mushroom freezing have been reported (Anon, 1964; Anon, 1971a); the major disadvantage of cryogenic freezing methods is the high cost.

2.5.3 Dehydration

Dehydrated foods have several advantages, most important of which are that they can be packaged, stored and transported more cheaply because of reductions in both weight and bulk. Mushrooms, which are usually about 90% water, are a product for which dehydration is a logical consideration.

Dehydrated mushrooms enjoy a limited commercial market in North America. They are used in various forms (powdered, kibbled, sliced etc.) primarily in soup and sauce mixes. According to some sources, the freeze dried product is the preferred form in that it has good aroma and flavor with an attractive color (Fang et al., 1971; Komanowsky et al., 1970). Small volumes of cultivated mushrooms are freeze dried on a commercial basis in the U.S. (Fang et al., 1971). A procedure used in a commercial mushroom freeze drying plant has been described by Luh and Woodroof (1975).

Hot air dehydration, on the other hand, is the most popular method for preserving various wild and cultivated

mushrooms in other countries (Komanowsky et al., 1970). According to Luh and Woodroof (1975) species of Boletus are the world's most commonly available variety of dried mushrooms; these are always collected in the wild. In Japan, Shiitake is grown commercially on a large scale, and up until recently, was mainly marketed in dried form (Fujimoto et al., 1972). Similarly, Chinese Ear Fungus is another very popular dried mushroom throughout Asia (Gray, 1970). Home sun drying of wild edible mushrooms is a common practice throughout Eastern Europe (Jelen, 1978).

Once again, the issue of whether to blanch prior to processing is an important question. Mushrooms which are not blanched before dehydration are subject to intense discoloration by browning both during drying and upon rehydration. The problem has been studied by several researchers.

An early work by Cruess and Mrak (1942) concluded that while dehydrated mushrooms which have been blanched are less attractive in appearance, flavor is superior to unblanched samples. On the other hand, some authors have reported loss of flavor on blanching (Luh and Woodroof, 1975; Ciobanu et al., 1976).

Fang et al. (1971) compared the final product quality of freeze dried mushrooms as it is affected by three pretreatment methods. These were:

a. a sodium metabisulfite solution (200 ppm) dip for 10 minutes,

b. a sodium chloride (2% solution) dip for 10 minutes,

c. water blanching (100°C) for 2 minutes followed by evaporative cooling.

He reported that the blanching treatment resulted in the best rehydration capacity, color (Hunter lightness value) and texture (Lee Kramer score) of the rehydrated products. Taste panel scores for flavor and aroma of the blanched samples, however, were not nearly as high as those for sulfited samples.

Komanowsky et al. (1970) investigated several additives for their effect on color preservation (i.e. citric acid, sodium chloride, ascorbic acid, EDTA, sodium acid pyrophosphate and sodium bisulfite). They found that only sodium bisulfite (less than 100 ppm) decreased discoloration during drying. Luh and Eidels (1969) observed similar results in that samples treated with 1% sodium bisulfite had substantially better color on rehydration.

While enzymatic activity is greatly retarded in an unblanched dehydrated food, it does once again become a problem when the material is rehydrated. Mushrooms which have not been blanched prior to dehydration are subject to rapid enzymatic browning at this time because of high levels of PPO. Metabisulfite and sulfur dioxide are compounds which prevent enzymatic browning by reducing products of the reaction back to their parent molecules whilst themselves being consumed in the reaction (Walker, 1977). Thus, if proper sulfiting techniques are employed, the problem of

browning during dehydration and upon rehydration can be eliminated.

The texture and flavor of dried mushroom products are, of course, also of concern. While little information is available on this aspect with specific reference to mushrooms, it is known that final moisture content and drying temperature affect both flavor and texture. Komanowsky et al. (1970) have shown that hot air dried mushrooms with 2% residual moisture were tougher on reconstitution than those with 7%, and as well, had a "somewhat burnt or bitter flavor", especially when dried at temperatures of over 80°C. Bartholomai et al. (1975) reported that the preparatory freezing treatment has an important effect on the flavor retention of a freeze dried mushroom extract. Their results showed that with rapid freezing (i.e. liquid nitrogen, -196°C for 2 minutes) only 53% of the volatiles were retained after lypophilization whereas samples frozen in still air (-40°C) retained 89%.

According to Tanaka et al. (1976), the texture of rehydrated freeze dried mushrooms is considered closest to the fresh product; air dried samples are tougher. Yeh (1971) investigated the texture of blanched and unblanched samples of air dried and freeze dried mushrooms after rehydration. The lowest shear press values were obtained for unblanched freeze dried samples, but unblanched air dried samples were more tender than the blanched freeze dried ones. Thus, it would appear that blanching also has the effect of

increasing the toughness of the final product regardless of the method of drying. Its use as a pretreatment should be assessed for this as well as previous reasons.

There are several types of drying methods which can be used for dehydration of vegetables. These include: hot air drying, drum drying, fluidized bed drying, vacuum drying and freeze drying. Of these, three have been used for mushrooms.

Freeze drying, as has already been mentioned, has some merits over other methods. However, a successful method for the production of an air dried product as an alternative to the freeze dried one has been reported in the U.S. (Anon, 1968a; Anon, 1968b; Anon, 1971b). In this process, mushrooms are first dipped for 10 minutes in a chlorine solution (500 ppm) to reduce microbial populations and then sulfited to inhibit browning. Tray drying is carried out in two stages; 3 hours at 43°C during the first stage and 1 hour at 79°C in the second and final stage. Moisture content of the final product should be in the range of 4 to 7%. Taste panels were used to establish shelf stability for periods of up to seven months; the product was rated as having acceptable flavor, color and texture. Flavor is said to be at least equal to, if not better than, that of a freeze dried product. Komanowsky et al. (1966) developed a drum dried mushroom powder in their laboratories that is also claimed to be equal or superior to a freeze dried mushroom powder.

2.5.4 Novel Processing Methods

A few enterprising individuals have investigated a number of novel processing methods for mushrooms.

Because of the relatively high protein quality of mushrooms and good nutrient composition generally, some workers have suggested the use of mushrooms for meat substitutes and/or extenders. For example, Janes and Ziemba (1972) reported on the use of mushrooms in fabricating one of five 'Patty Pleasers' marketed at that time for the Hotel-Restaurant Industrial (HRI) trade in the United States. Similarly, a method was developed whereby mushrooms are ground and cooked into patties directly; these were reported to have a texture similar to ground beef (Anon, 1967). Mushrooms have also been suggested as a raw material for sausage production (Uzunov and Colova, 1972).

Among the convenience type products, mushrooms have been proposed for such uses as:

a. a frozen French-fried product (Gormley, 1973),
b. chips prepared from deep-fat fried mushrooms (Greenspoon et al., 1964),
c. a candied product (in which mushrooms are soaked in a citric acid and sugar syrup before being cooked) (Hirota, 1973).

2.5.5 Research on the Oyster Mushroom

Industrial research and commercial processing of the Oyster Mushroom is advanced in Czechoslovakia. At 'Vyzkumny ustav lihovarov a konzervarni' in Bratislava,

Czechoslovakia, large amounts of Oyster Mushrooms are cabinet dried and some canned products are also being produced (Weijer, 1978). It is likely, however, that these operations are proceeding on an industrial trial and error basis as no published data have been released.

To the author's knowledge only two papers have been published on the processing potential of the Oyster Mushroom. Both were concerned mainly with Pleurotus ostreatus with little emphasis on the Pleurotus florida variety.

The first one, by Andreotti et al. (1975), consisted of chemical analyses, storability experiments, and cursory observations on suitability of Pleurotus ostreatus for drying, canning and freezing. Briefly, their conclusion was that Pleurotus ostreatus is suitable for canning and freezing but not for drying. The information in this paper is primarily speculative; very few of the tests were based on objective or subjective measurements.

The second paper, a work by Gormley and O'Riordain in 1976, was better documented. Investigations consisted of shelf-life tests, freezing, and dehydration experiments on Pleurotus ostreatus, and in a few cases on Pleurotus florida. They concluded that the Oyster Mushroom "...is not well suited to freezing" and that although they "...lend themselves to dehydration...there are some problems associated with the dried and reconstituted products". Specific reference to the results reported by these authors

will be made in the experimental sections which follow.

The conclusions drawn by the above authors are somewhat contradictory. It is hoped that the present work will clarify the situation and provide some perspective on the advantages and disadvantages of Pleurotus florida for processing.

3. EXPERIMENTAL MATERIALS AND TASTE PANEL SELECTION METHODS

3.1 INTRODUCTION

The species Pleurotus florida is known to have certain advantages over Pleurotus ostreatus from a cultivation standpoint. These are: greater ease in the mycelial growth phase, a fructification temperature requirement in the range of normal room temperatures, higher yields per unit area with good consistency, and a lower proportion of stems to caps (Zadrazil and Schneidereit, 1972). It was primarily for the first two reasons that Pleurotus florida was utilized for all experimental work in this project.

The first section in this chapter describes the method which was used to grow the mushrooms. Photographs taken at various stages are included in order that the reader can better visualize the production process. A section on yield data from first, second and third flush crops and measurements on the average proportion of stems to caps follows. Complete proximate analyses were performed on cumulatively prepared samples of stems and caps from first and second flushes; these results are reported in the fourth section. In the last section, the methods used for taste panel screening are described.

All experiments in the project utilized Pleurotus florida cultivated by the method described below. In cases where Agaricus bisporus was used for comparative purposes, the material (supplier: Prairie Mushrooms, Edmonton) was

purchased at local supermarkets as required.

3.2 CULTIVATION METHOD FOR PLEUROTUS FLORIDA

All cultivation work described below was carried out by the author using existing facilities of the Department of Genetics, University of Alberta.

3.2.1 Preparation of Spawn

Pleurotus florida spawn (Plate 1) was prepared by the following method:

a. Wheat kernels were cooked in distilled water (1:1.5 w/v) at 100°C for approximately 45 minutes and dispensed into 1 liter flasks.

b. The flasks were covered and autoclaved for 90 or 150 minutes at 121°C using 16 lbs pressure (The initial supply of wheat required only 90 minutes; for subsequent batches of wheat, 150 minutes had to be employed for adequate sterilization because of a greater degree of contamination).

c. Cooled flasks were inoculated aseptically with master culture prepared from pure cultures of *Pleurotus florida* supplied kindly by Dr. J. Weijer, Department of Genetics, University of Alberta.

d. The inoculated flasks were incubated 10-14 days at approximately 25°C and stored under refrigeration (4°C) until needed.

3.2.2 Substrate Preparation

Wheat straw for use as substrate was first chopped (approximately 2.5 - 5.0 cm length) using a silage maker (Plates 2 and 3). It was then soaked in an excess of water (Plate 4) to obtain a final moisture content of approximately 70%, bagged (about 2.5 kg) in large polyethylene bags and pasteurized (60°C for 72 hours) in a large incubator.

3.2.3 Mycelial Phase

Cooled pasteurized substrate was inoculated with approximately 2% (w/w) spawn. Individual bags of inoculated straw were then pressed into wooden boxes (volume: 0.28 cubic meters) and placed into a dark chamber with 50% R.H. at 25°C (Plate 5). After about 14 days incubation dense mycelial 'blocks' were formed. Plate 6 illustrates the appearance of these blocks at the completion of this phase.

3.2.4 Fructification Phase

The unmolded straw blocks were stacked (3 to 4 high) in a high humidity chamber (75-80% R.H.); temperature was normally 22 - 25°C. Continuous lighting was provided with fluorescent tubes placed in succession about 150 cm from the floor of the chamber (Plate 7). Fruit bodies (Plates 8 and 9) developed after 7-10 days.

It was found that successive crops could be produced if the straw blocks were cleaned of all debris, moistened and

put back into plastic bags in the dark chamber for a 10 day rest period before receiving another light phase. Mold contaminants were sometimes a problem, particularly with third flush crops.

Plate 1 *Pleurotus florida* spawn.

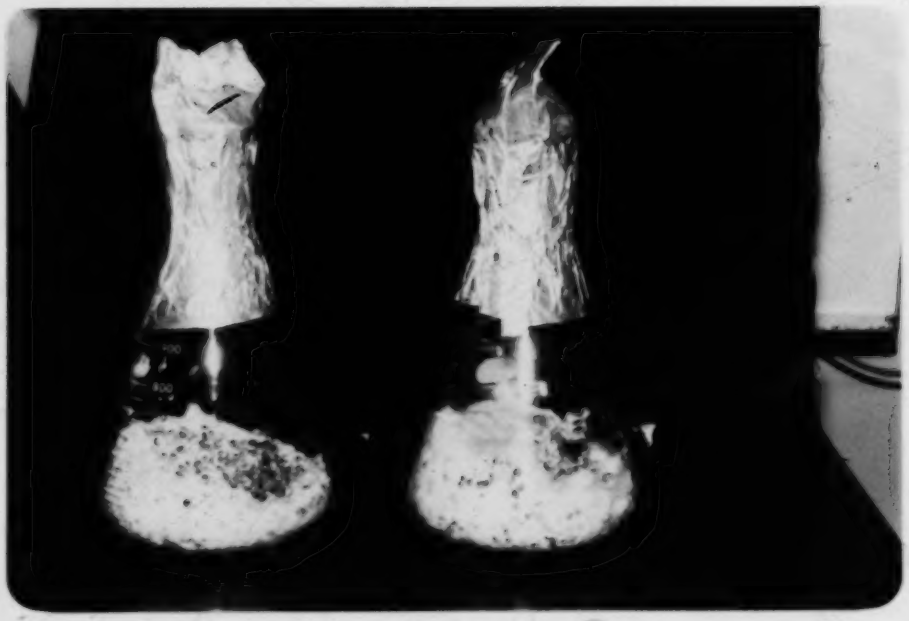


Plate 2 Apparatus for chopping straw substrate.

Plate 3 Appearance of substrate before and after chopping.

Plate 4 Substrate soaking operation.

Plate 5 Inoculated substrate during the mycelial growth phase.

Plate 6 Appearance of straw 'blocks' at completion of mycelial growth phase.

Plate 7 Light chamber for fructification phase.

Plate 8 *Pleurotus florida* fruit bodies (top view).

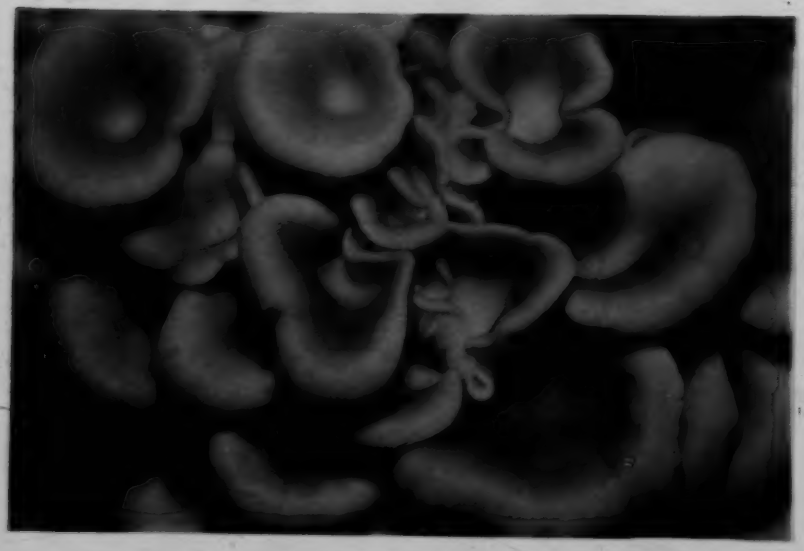


Plate 9 *Pleurotus florida* fruit bodies (side view).

3.3 YIELD DATA

3.3.1 Methodology

The weights of fresh mushrooms harvested from first, second and third flush crops were recorded for 10 cultivation runs. In order to assure as much uniformity as possible, the entire crop of fruit bodies was removed by cutting at the point of contact with the straw. The proportion of stems to caps was also determined (for first flush crops only) by separating the two portions and weighing each respectively. Yields were expressed on the basis of kilograms of fresh mushrooms harvested per cubic meter of substrate. The average weight of mushrooms actually harvested per cultivation run was about 2 kilograms.

3.3.2 Results

Average yields for first, second and third flushes are given in Table 4. It is apparent that with succesive crops, yield decreased in an approximately linear fashion. Zadrazil (1973b) reported that second flush crops of *Pleurotus florida* produced only 25% the weight of mushrooms when compared to first flush yields. The differences between his results and those reported in Table 4 could possibly be explained by cultural techniques. Hashimoto and Takahashi (1976) found second flush yields of *Pleurotus ostreatus* to be about 50% of first flush yields. These findings agree well with the data reported in Table 4 for *Pleurotus*

Table 4 Yield data for first, second and third flush crops of *Pleurotus florida* (fresh weight).

Sample	# of Trials	Maximum (kg/m^3)	Minimum (kg/m^3)	Mean (kg/m^3)	Std. dev.
1st flush	10	17.6	11.0	14.5	2.1
2nd flush	10	9.0	7.1	8.0	0.5
3rd flush	10	4.5	3.0	3.8	0.4

florida.

Table 5 shows the results obtained for yields of caps and stems. Under the growth conditions used in this experiment roughly 20% of the total mass harvested from a first flush crop was contributed by the stem portion of the mushroom. Kalberer and Vogel (1974) reported a stem proportion of 38-51% for Pleurotus ostreatus. Thus, the results of this experiment appear to substantiate the claim made by Zadrazil and Schneidereit (1972) that the Pleurotus florida species has a lower proportion of stems. It should also be noted, however, that the level of light provided will influence the extent of stem development.

3.4 PROXIMATE ANALYSES

The stem and cap portions of both first and second flushes were analyzed for moisture, protein, fat, fiber, ash and reducing sugars.

Records for different cultivations runs, kept over a period of several months, revealed that percent total solids (%T.S.) ranged from approximately 7 to 12%. For this reason, batches of material, from five separate cultivation runs, were pooled to obtain more representative values for the proximate analyses.

Moisture content of the fresh materials was first determined and then 100 gram samples of each were freeze dried, vacuum packaged and stored at -30°C. The process was repeated four more times and moisture of the composite

Table 5 Weight proportion of caps to stems in *Pleurotus florida* fruit bodies (first flush crop).

Sample	# of Trials	Maximum %	Minimum %	Mean %	Std. dev.
Stems	10	30.04	17.03	21.18	4.25
Caps	10	82.97	69.96	78.82	4.25

samples was determined. The freeze dried materials were ground and the powders obtained used directly for protein, fat, ash, reducing sugars and fiber analyses. Results for these fractions are expressed on a dry weight basis. All analyses were performed in duplicate, unless otherwise specified. Statistical significance of the differences between the first and second flushes of Pleurotus florida stems and caps was evaluated by the t-test.

3.4.1 Methodology

a. Moisture Content

Moisture content was determined using the routine vacuum oven method. Fresh materials were finely chopped, mixed and a 10 g subsample removed. The freeze dried materials were powdered and a 5 g subsample used. The oven was set at 60°C with a 29" Hg vacuum for 48 hours. Each determination was done in triplicate.

b. Protein

Total nitrogen was determined using the micro-Kjeldahl method (A.O.A.C., 1975); all samples were 0.15 g in size. Percent crude protein was calculated using % nitrogen X 6.25 as the conversion factor.

c. Fat

a) Crude fat content was determined by the Soxhlet method (A.O.A.C., 1975).

Approximately 5 g of powdered materials were extracted for 8 hours with petroleum ether. The solvent was removed by roto-evaporation and the lipid dried to constant weight at room temperature in a desiccator.

b) Because of the possible existence of both polar and non-polar lipids, an alternative method was also used to measure fat content. The method chosen was chloroform/methanol extraction (Bowyer and King, 1977). Only first flush caps were tested.

d. Reducing Sugars

These analyses were conducted by the Food Laboratory, (O.S. Longman Bldg., Edmonton, Alberta) using a modified Munsen Walker Method (A.O.A.C., 1975).

e. Crude Fiber

Crude fiber was determined by the Official Method (A.O.A.C., 1975). This method measures crude fiber by incineration of the dried residue after digestion with dilute sulfuric acid and sodium hydroxide.

f. Ash

Ash was determined by the method outlined in A.O.A.C. (1975). Samples (2 g) were ashed at 550°C to constant weight (approximately 5-6 hours). Quadruplicate determinations were made.

3.4.2 Results

Results for the proximate analyses are given in Table 6. As expected, the two portions of the mushroom differed in their composition. Furthermore, differences were also observed between first and second flushes.

Moisture content, when determined as an average value for five runs, varied little between the four materials and was in the range of 91%. Thus, when moisture is measured in this way, the results appear to be similar regardless of whether one is dealing with caps or stems, first or second flush. This is in contrast to the variability observed for individual cultivation runs. Other differences were, however, apparent.

On a dry weight basis, crude protein values ranged from 41% in first flush caps to 18% in second flush stems. Caps contained the highest amounts of protein, 41% and 36% for first and second flushes respectively. These values were significantly higher than those found for the corresponding stems ($p \leq 0.01$). In addition, crude fiber was lower and ash higher in caps than in stems for a given flush ($p \leq 0.05$). Lower values for crude fiber in the caps agreed with their observed higher tenderness.

Since the method of analysis used to determine carbohydrate measured only reducing sugars, the possibility exists that considerable amounts of carbohydrate went undetected. According to Holtz (1971), approximately 70% of the carbohydrate of Agaricus bisporus is contributed by

Table 6 Proximate composition of *Pleurotus florida* caps and stems from first and second flushes.

Sample	Water % fresh weight	Crude protein	Crude fat	Sugars (red.)	Crude fiber	Ash	Unknown
			% dry weight				
1st flush caps	91.92	40.93	2.12[1] 6.30[2]	33.84	9.96	8.76	4.39
1st flush stems	91.26	25.09	1.40	25.07	13.48	7.69	27.27
2nd flush caps	91.82	36.31	2.24	41.72	11.19	7.06	1.48
2nd flush stems	91.51	18.36	1.18	35.27	14.72	5.36	25.11

[1] Petroleum ether extraction

[2] Chloroform/methanol extraction

mannitol (non-reducing sugar alcohol). The total of the results reported in Table 6 is close to 100% for first and second flush caps, suggesting that their proximate compositions have been fairly well described. Results for stems, on the other hand, indicate an unidentified portion of approximately 25% in both cases. It is speculated that either the carbohydrate fraction of these two stems samples was underestimated because of a high presence of non-reducing sugars (or other sugar compounds) or that the methods of analysis did not give a complete determination for the other components. The latter might to be due to various possible physical or chemical causes. In general, carbohydrate is an important component representing one third or more of the dry matter.

The main differences found between first and second flush crops for a given portion of the mushroom were:

a. An increase in the amount of fiber from the first to the second flush ($p \leq 0.05$),

b. A decrease in protein and ash contents from the first to the second flush ($p \leq 0.05$ and $p \leq 0.01$ respectively).

If one uses protein content as a gauge for measuring nutritional quality, then it could be said that mushrooms from first flush crops score the highest. A similar trend is also apparent for caps versus stems. Consequently, it is necessary to consider both of these factors in reporting the protein content of this mushroom. In fact, this observation

may explain the somewhat lower values quoted in the literature for the Oyster Mushroom. For example, Kalberer and Kunsch (1974) estimated the dry weight protein content of Pleurotus ostreatus to be 30%; Andreotti et al. (1975) reported values in the range of 20%. The protein content determined in the present work for first flush caps of Pleurotus florida, (41%), is comparable to the value determined for Agaricus bisporus by Kurtzman, (1975).

It is of special interest to note the large difference observed in the fat analyses depending on which of the two methods was used. The application of the chloroform/methanol extraction method recovered approximately 3 times more lipid. This effect is most likely due to the presence of large quantities of polar lipids (eg. phospholipids). Lee and Mattick (1961) reported the same effect for lipid analyses of peas. Further discussion on the lipid fraction will be presented in a later section (see section 5.4).

3.5 TASTE PANEL SCREENING

As it was realized that sensory evaluation would form an integral part of the evaluation of the processing potential of Pleurotus florida, it became important to consider various aspects of its use. Taste panel evaluations of product quality parameters frequently have inherent limitations, but often no other suitable means of assessment is available. In the present work, attempts were made to increase the efficiency of the panel as well as to chose

testing procedures which would effectively serve the purpose.

It is believed that 'trained' judges are several times more efficient than untrained counterparts in that their sensitivity is higher and their ability to differentiate improved (Larmond, 1970). According to Zook and Wessman (1977), the first step is screening; if panels are to be used for descriptive work then training sessions must also be conducted. The panel chosen for use in this project was screened (see below) but cannot be considered trained. Since the aim of most tests was to assess sample acceptability, training per se was not felt to be appropriate.

Certain types of testing procedures have been demonstrated to be more efficient than others. For instance, the variables multiple comparison test (in which each sample is scored or rated) is several times more powerful than duo-trio or triangle tests (Kramer, 1973). Thus, the approach of this work was to utilize, wherever possible, the aforementioned test. Common problems (i.e. lack of replicates, panelist availability, variability in panelist's judgements etc.), however, placed limitations on the strength of the conclusions drawn.

3.5.1 Selection of Panelists

Sixteen staff and student members of the Department of Food Science, University of Alberta, participated in the screening sessions. In all, seven sessions were conducted to

choose the final panel consisting of eight judges. These individuals were then asked to assist in sensory evaluations throughout the project.

3.5.2 Ability to Distinguish Different Mushroom Flavors

The flavor of different mushroom species is usually quite unique although some similarities are also apparent. For this reason, it was important that the individuals chosen for the panel have a sense of acuity for mushroom flavors; the ability to distinguish these was tested by the following procedure.

A mildly flavored cream of mushroom soup base was prepared and concentrated artifical flavor extracts (1 g) of either 'H&R Champignon' US-93900 or 'H&R Steinpilz' US-49223 (supplier: Haarmann & Reimer, Division of Bayer (Canada) Inc.) were added. Panelists were asked to identify the odd sample in a standard triangle test (Larmond, 1970). Two replicates of this test were performed.

3.5.3 Ability to Distinguish Mushroom Flavor Intensity

The ability to distinguish intensity of a mushroom flavor was also felt to be an important criterion. The following test was employed to achieve this end.

Five concentrations of cream of mushroom soup bases were formulated using 0, 1, 2, 3, and 4 parts of the 'H&R Steinpilz' extract. Panelists were asked to rank the samples according to increasing mushroom flavor intensity. Three

replicates of this test were used.

3.5.4 Ability to Distinguish Textural Differences

An indication that changes in the texture of mushrooms could be detected by the panelists was felt to be crucial. The following procedure was used to evaluate this ability.

One batch of fresh mushrooms (*Agaricus bisporus*) was steam blanched for 3 minutes, another for 10 minutes. (The sample blanched for 10 minutes was much firmer because of a greater moisture loss). Panelists were given one of each sample and asked to indicate which had the firmer texture; they were also asked to indicate which one they preferred. The second question was included because there was no reason to presuppose that one sample would be more acceptable than the other. In this way, some information on the preference for mushroom texture was gained. Two replicates were used.

3.5.5 Choice of the Final Taste Panel

Results from the seven screening sessions were reviewed carefully and eight individuals were chosen based on their relative success in scoring correctly. All of those selected were successful in chosing between different mushroom flavors in both tests, were able to correctly order samples for mushroom flavor intensity in at least two out of three trials and could distinguish textural differences accurately. It is of interest to note that no preference pattern for mushroom texture was observed. Roughly half of

the group preferred the less firm sample and half the more firm sample.

4. BLANCHING STUDY

4.1 INTRODUCTION

The importance of blanching as a pretreatment for further processing of mushrooms has already been discussed. The main problem it poses is loss in product weight. A considerable amount of effort has been expended in assessing these losses for *Agaricus bisporus* and suggesting means to reduce them (McArdle and Curwen, 1962; Beelman *et al.*, 1973; Beelman and McArdle, 1975; Beelman *et al.*, 1976; Eby *et al.*, 1977). Very little is known, however, about the effect of blanching on *Pleurotus florida*. The questions which this study attempted to answer were as follows:

a. What is the time required for adequate blanching treatment of *Pleurotus florida*?

b. How do steam and water blanching differ in their effect on these mushrooms?

c. Do stems and caps behave similarly with regard to blanching losses?

d. Are blanching effects the same for mushrooms from different flushes?

e. How does *Pleurotus florida* compare with *Agaricus bisporus* in terms of blanching shrinkage?

f. Does post harvest storage decrease blanching losses?

g. What is the extent of shrinkage in terms of volume change?

4.2 MATERIALS AND TESTING METHODS

4.2.1 Peroxidase Test

The peroxidase test is frequently used to assess the adequacy of a blanching treatment. In order to determine the time necessary for destruction of this enzyme in Pleurotus florida, the rapid method of Fang et al. (1976b) was used. Mushrooms (50 g sample size) were steam blanched for 1 to 5 minutes, cooled in running water and drained before being tested for peroxidase activity. The minimum time required for a negative test result, and thus for adequate blanching, was determined and used for all blanching experiments in this section of the work.

4.2.2 Blanching Tests

Water and steam blanching tests (see below) were conducted using three cultivation runs of each of the following materials:

first flush Pleurotus florida caps,
first flush Pleurotus florida stems,
second flush Pleurotus florida caps,
second flush Pleurotus florida stems.

Freshly harvested mushrooms (50 g sample size) were used. Similarly, these blanching tests were also run on three lots of Agaricus bisporus for comparative purposes. Average values for percent shrinkage and standard error were calculated from the data recorded on weight loss.

In addition to weight loss, the total solids (T.S.)

content before and after blanching was also determined. Accordingly, moisture was measured by the method previously outlined (page 60).

The effect of post harvest storage on blanching losses was examined for first flush caps only. One batch was steam blanched (see below) immediately after harvest, the other was stored for 24 hours at 4°C before receiving an identical blanching treatment. Three cultivation runs were used. Weight losses were recorded for the calculation of percent shrinkage.

All results were analyzed by the Analysis of Variance (ANOVA) test.

4.2.2.1 Blanching Procedures

Preweighed materials (50 g) for steam blanching were placed into standard household sieves and heated for the specified time in a steam table at 95°C. Following blanching, they were removed, cooled in cold running water for 30 seconds, drained 2 minutes, blotted on a single paper towel (1 minute) and reweighed. All determinations were in triplicate.

Preweighed materials (50 g) for water blanching were placed in beakers of distilled water (1:10 w/v), held at 95°C in a water bath. These were left undisturbed until the original water temperature was regained (about 1 - 1.5 minutes) and then held for 3 minutes. After removal they were treated in the same manner as the steam blanched

materials before being reweighed. All determinations were in triplicate.

4.2.2.2 Solids Mass Balance Experiment

Solids mass balances were conducted in duplicate using stems and caps of *Pleurotus florida* from first and second flushes. All materials (from one cultivation run only) were stored 24 hours at 4°C before use. Again, *Agaricus bisporus* was used for comparative purposes. Preweighed tin cans (306 X 411) were filled with 100 grams of each mushroom material. Distilled water at 25°C was added (approximately 275 - 300 g) and the tins reweighed to determine the weight of water. Sealed tins, fitted with thermocouples, were immersed in a jacketed steam kettle containing water at 100°C; temperature in the center of the can was monitored on a strip chart recorder such that each tin received a heat treatment equivalent to 3 minutes at 95°C. Upon removal, the tins were cooled immediately in ice water, opened, and the two fractions carefully separated and weighed. Total solids analyses were performed on both fractions and the results recorded.

4.2.2.3 Measurement of Volume Shrinkage

Volume shrinkage of the cap and stem portions of *Pleurotus florida* (first flush crop) and *Agaricus bisporus* was measured in the following way. Samples of each material were finely chopped and dispensed uniformly into test tubes

to a level of approximately 10 centimeters. The tubes were capped, put on a mechanical shaker for 5 minutes, tapped down and placed into a beaker of water at 100°C for 5 minutes. After removal, they were cooled for 5 minutes, put back on the shaker for an additional 5 minutes, tapped down again and the final heights determined. The difference between the height of the column of mushrooms before and after blanching was used as a measure of the volume shrinkage. Five replicates were used for each material.

4.3 RESULTS AND DISCUSSION

4.3.1 Blanching Time

Data obtained from the peroxidase test are given in Table 7. The results indicated that a blanching time of at least 3 minutes in steam was necessary to inactivate the peroxidase enzyme. Hence, all experiments on water and steam blanching were conducted using 3 minutes as the blanching time.

4.3.2 Water and Steam Blanching Tests

Data from T.S. measurements and weight loss determinations are shown in Table 8. The values reported here are the calculated averages from three cultivation runs with three replicates per run and are therefore fairly representative.

With respect to changes in total solids, the trend was generally to an increase when steam blanching was used and a

Table 7 Test for peroxidase activity in *Pleurotus florida* after steam blanching treatments.

Blanching time (minutes)	Peroxidase activity
1	positive
1.5	positive
2	positive
2.5	positive
3	negative
4	negative
5	negative

Table 8 Steam and water blanching of *Pleurotus florida* and *Agaricus bisporus*: effect on % T.S. and weight loss (mean ± standard error).

	Initial	After Steam Blanching		After Water Blanching	
	T.S. (%)	T.S. (%)	wt loss (%)	T.S. (%)	wt loss (%)
P. florida (1st flush)					
caps	9.1 ±0.2	10.0 ±0.2	19.0 ±0.9	7.0 ±0.3	9.8 ±2.1
stems	9.0 ±0.2	10.6 ±0.2	17.0 ±1.2	8.0 ±0.3	12.5 ±2.1
P. florida (2nd flush)					
caps	8.3 ±0.2	9.2 ±0.0	13.8 ±1.2	7.3 ±0.2	2.5 ±1.5
stems	10.0 ±0.4	11.4 ±0.3	3.1 ±2.1	8.9 ±0.5	3.8 ±2.5
A. bisporus					
caps	8.1 ±0.4	8.2 ±0.2	24.6 ±1.5	8.3 ±0.2	27.8 ±2.5
stems	8.2 ±0.1	7.9 ±0.4	15.4 ±1.5	7.4 ±0.2	16.0 ±1.0

decrease with water blanching. This implied that with steam blanching moisture was removed but that with water blanching either solids were lost or moisture gained or both. It was primarily for this reason that the solids mass balance experiment was conducted; the objective was to determine which of the effects was responsible (see section 4.3.4).

Results of the statistical analysis of the data on weight loss are presented in six tables which can be found in Appendix A. The findings can be summarized as follows:

a. Pleurotus florida lost significantly less weight during blanching than Agaricus bisporus.

b. Water blanching of Pleurotus florida resulted in significantly lower weight losses than steam blanching; a similar effect was not observed for Agaricus bisporus.

c. The stems of Agaricus bisporus and Pleurotus florida lost significantly less weight during blanching than their respective caps.

d. Second flush Pleurotus florida lost significantly less weight during blanching than first flush Pleurotus florida.

4.3.3 Effect of Post Harvest Storage

An effect of storage has been demonstrated on the blanching shrinkage of Agaricus bisporus (Beelman et al., 1973). Hence, the results reported for Agaricus bisporus and Pleurotus florida in Table 8 may not give an accurate comparison because one material (Agaricus bisporus) was

stored and the other (Pleurotus florida) was not.

When first flush Pleurotus florida caps were stored for 24 hours at 4°C and compared to freshly harvested Pleurotus florida caps for weight shrinkage by steam blanching, results of 13.5% and 19.0% respectively were obtained. Therefore, more representative values for the average shrinkage of the two mushrooms would be 13.5% for Pleurotus florida and 25.0% for Agaricus bisporus; these means are significantly different at the 1% level. A similar difference was also apparent in the mass balance experiment where samples of Pleurotus florida stored 24 hour post harvest were again used (see Table 9). McArdle and Curwen (1962) also found the average shrinkage of Agaricus bisporus after blanching to be 25%.

According to the work of Eby et al. (1977), there is a positive relationship between the level of free amino acids in mushroom tissue and water-holding capacity (WHC). They further state: "The rise in free amino acid content is probably due to proteolytic enzyme activity which is known to increase during postharvest storage of mushrooms." It may be possible that freshly harvested Pleurotus florida fruit bodies contain a higher initial level of free amino acids than Agaricus bisporus and that these levels are enhanced by post harvest storage. This could explain the observation of a higher WHC in Pleurotus florida. It is also likely that the high fiber content may play an additional role in increasing the WHC, especially in stems and/or second flush

mushrooms.

4.3.4 Effect of Water Blanching on Solids and Weight Losses

The results obtained from the solids mass balance water blanching experiment are given in Table 9. From these results, it is apparent that a possibly undesirable solids loss will occur. Water soluble carbohydrates and minerals are suspected to be the main components responsible. Approximately 30% of the solids in *Pleurotus florida* were leached, and even more, 41%, in the case of *Agaricus bisporus*. There was essentially no difference between the caps and stems of *Pleurotus florida* in terms of the extent of solids losses, but differences were again observed in percent weight loss (i.e. stems shrank less than caps). Again, *Pleurotus florida* showed significantly lower weight losses than *Agaricus bisporus*.

4.3.5 Effect of Blanching on Volume Shrinkage

Table 10 presents the results obtained for the effect of blanching on volume changes of *Pleurotus florida* and *Agaricus bisporus*. It was expected that *Pleurotus florida* caps would shrink substantially more in terms of volume because of the large exposed gill area. The results showed, however, that although the percent volume change was greater for caps of *Pleurotus florida* than those of *Agaricus bisporus*, the difference (about 7%) was not as large as anticipated. It is also interesting to note that the stems

Table 9 Solids and total weight losses of *Pleurotus florida* and *Agaricus bisporus* after water blanching.

Sample	Initial T.S. %	Solids loss* %	Water loss/ gain %	Total weight loss %
Pleurotus florida (1st flush)				
caps	8.3	32.5	-8.2	11.3
stems	9.0	30.0	-7.4	10.3
Pleurotus florida (2nd flush)				
caps	8.0	31.2	-6.0	8.7
stems	8.2	32.1	+3.3	0.2
Agaricus bisporus				
caps and stems	8.1	41.5	-15.9	21.3

* calculated on dry weight basis

Table 10 Volume shrinkage of *Pleurotus florida* and *Agaricus bisporus* as effected by 'blanching'.

Sample	Column Height before (cm)	Column Height after (cm)	Percent change
Pleurotus florida caps	8.9	4.3	62.6
Pleurotus florida stems	8.9	5.1	42.7
Agaricus bisporus caps	9.3	4.2	55.4
Agaricus bisporus stems	9.1	4.5	50.3

of Pleurotus florida decreased somewhat less in volume than stems of Agaricus bisporus. Stems of either mushroom did not experience as great a volume change as caps.

4.4 REMARKS

The experiments described in this chapter have demonstrated that Pleurotus florida is less subject to weight and solids losses during blanching than Agaricus bisporus. The influence of flush and the portion of the mushroom on shrinkage were demonstrated. Volume shrinkage was somewhat greater for Pleurotus florida than for Agaricus bisporus but not to a substantial level. These observations may have implications as to the suitability of Pleurotus florida for processing.

Blanching is a commonly used pretreatment for freezing, canning and drying preservation of most vegetables. In the case of mushrooms (Agaricus bisporus) this also applies. Its fundamental role in freezing preservation is to arrest enzymatic activity which can cause a decrease in product quality during storage. The situation for drying is somewhat more ambiguous because enzymatic activity is greatly retarded by the low water content and water activity. However, blanching is frequently applied for preservation of color both during drying and upon rehydration. In canning, the most important function of blanching is the removal of tissue gases and shrinkage of the product for satisfactory can fill. In the chapters which follow on the freezing,

canning and drying experiments with Pleurotus florida, the use of blanching and its effects on product quality are examined as a part of the overall evaluation of these processes.

5. FREEZING EXPERIMENTS

5.1 INTRODUCTION

A review of the literature on the freezing preservation of mushrooms (*Agaricus bisporus*) revealed that color retention is one of the biggest problems. Thus, the initial objective of this portion of the work was to ascertain how *Pleurotus florida* would compare to *Agaricus bisporus* in terms of the degree of enzymatic browning to which it is subject. Also to be considered was the need for blanching of *Pleurotus florida* in the maintenance of flavor components during frozen storage. The final concern was the effect of blanching treatment and/or the type of freezing regime on texture; drip loss was used as a measure of the extent of textural damage.

5.2 COMPARISON OF POLYPHENOLOXIDASE ACTIVITY AND SUBSTRATE LEVELS IN PLEUROTUS FLORIDA AND AGARICUS BISPORUS

Preliminary observations clearly indicated that *Pleurotus florida* was not subject to browning to nearly the same extent as *Agaricus bisporus*. Consequently, tests were performed to confirm this observation by comparing its PPO activity and substrate levels (phenolic compounds) to values determined for *Agaricus bisporus*.

5.2.1 Methodology

It is well known that a number of phenolic substances can react with PPO to form brown-colored end products. Thus, relative measures of PPO activities in Pleurotus florida and Agaricus bisporus were determined by the application of a phenolic substrate (4-methyl catechol) to the surface of the mushrooms and measurement of the resultant color. Similarly, the direct application of a pure PPO enzyme solution permitted the relative comparison of their tissue substrate concentrations, also based on color development.

Solutions of 0.01%, 0.10%, 0.50%, 1.0%, 2.0%, 3.0% and 10% 4-methyl catechol were prepared. Fresh samples (15 g) of sliced Agaricus bisporus and Pleurotus florida (cut into strips) were placed in petri dishes and wetted with 15 ml of the above solutions. Color, as lightness (L value), was determined after five minutes using a Hunter Colorimeter (HunterLab Color/Difference Meter D25-2). Untreated samples of the two mushrooms were used as controls. All determinations were in duplicate.

The levels of phenolic substrates were compared in samples of Pleurotus florida and Agaricus bisporus (prepared as above) by the application of 15 ml of an 0.1% solution of purified mushroom polyphenoloxidase (Tyrosinase; Sigma Chemical Co., Product #T-7755, lot #17C-9520). Lightness (L values on the Hunter Colorimeter) was measured at 1, 2, 3, 4 and 5 minutes. Care was taken to assure uniform positioning of the dishes in the instrument from one reading to the

next. Untreated samples of the two mushrooms were used as controls and all determinations were in duplicate.

5.2.2 Results and Discussion

From Figure 1 it can be seen that while the initial lightness readings of the untreated controls were very similar, a marked difference in browning was observed for the two mushrooms; as the concentration of 4-methyl catechol applied increased, the degree of tissue darkening was disproportionally lower for Pleurotus florida than for Agaricus bisporus. Lightness values of 19.4 and 46.9 were obtained for Agaricus bisporus and Pleurotus florida respectively with the most highly concentrated solution. These results suggest that the activity of PPO in Pleurotus florida is substantially lower than that found in Agaricus bisporus.

Regarding the levels of phenolic substrates present in the two mushrooms, Figure 2 shows the effect of the PPO contact time. Again, a distinct difference in the degree of browning was evident. The total decrease in lightness for Agaricus bisporus was 24.2 L units while that observed for Pleurotus florida was only 8.8 units. It therefore seems that in addition to a lower enzyme activity, Pleurotus florida does not have as high a concentration of substrate as Agaricus bisporus available for reaction with the enzyme.

Although much less sophisticated in methodological approach, the above observations are similar to those made

Figure 1 Relative activity of the enzyme polyphenoloxidase in Pleurotus florida and Agaricus bisporus as measured by the extent browning.

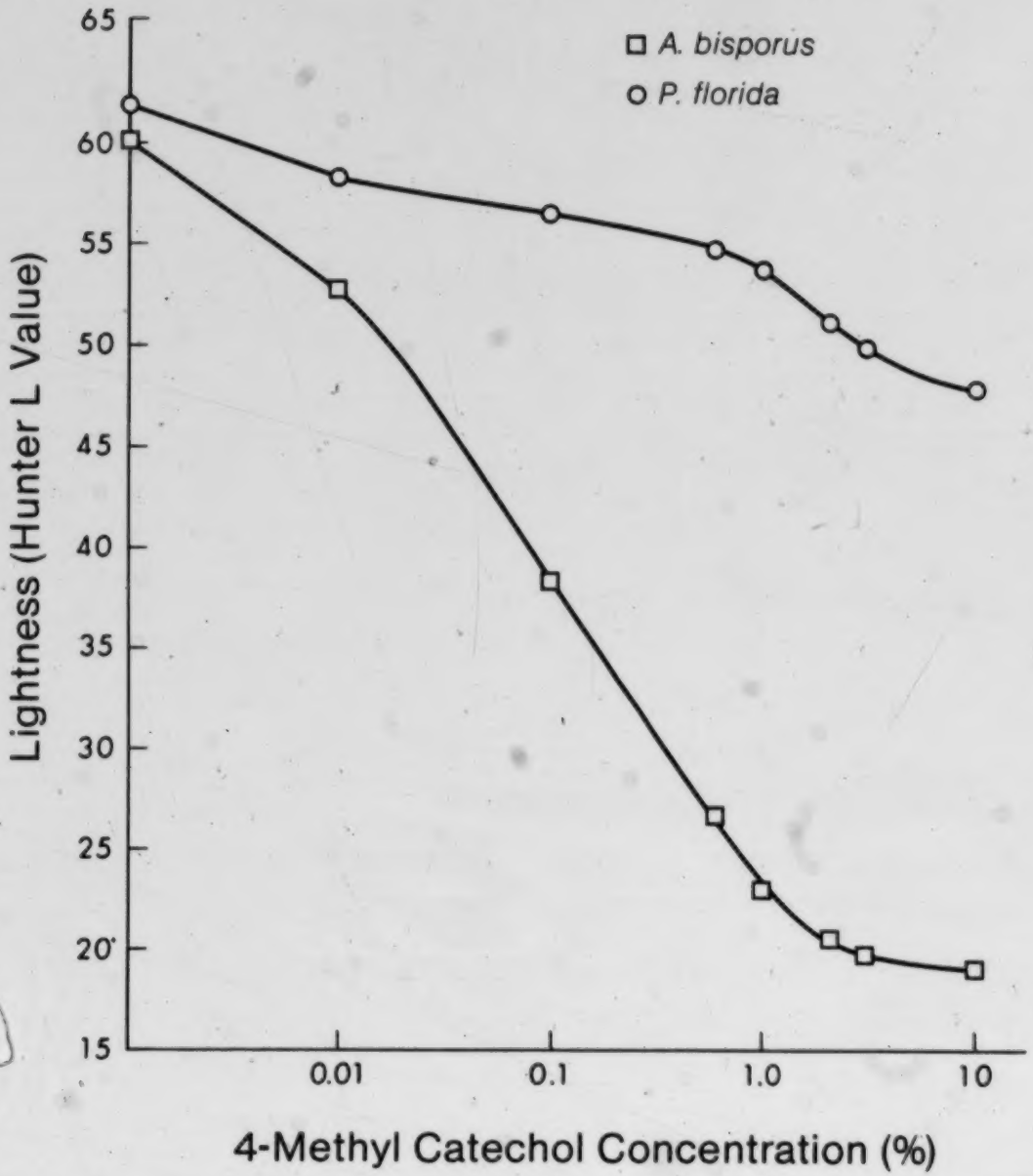
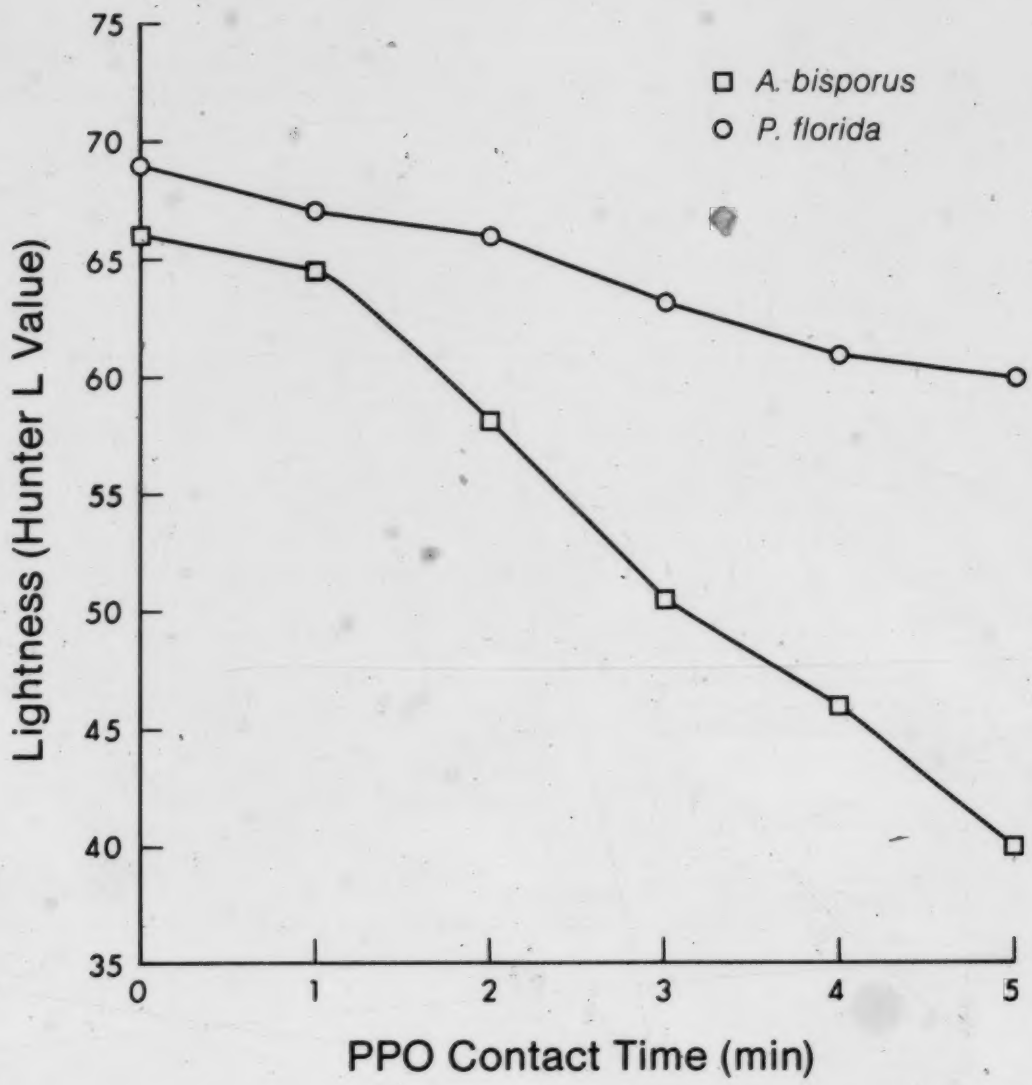


Figure 2 Relative activity of phenolic substrates in Pleurotus florida and Agaricus bisporus as measured by the rate of browning.



by Golan et al. (1977) for two avocado cultivars. They found that the rate of browning in one cultivar was much higher than in the other and that the effect was positively correlated to "...total phenols content and polyphenoloxidase activity".

Enzymatic browning in Agaricus bisporus is well documented and poses a serious problem. A considerable amount of effort has been expended to finding ways of reducing this problem either by physical or chemical methods. Therefore, the observation that Pleurotus florida is not subject to enzymatic browning to nearly the same extent as Agaricus bisporus could be an interesting and indeed important result.

5.3 EFFECT OF FREEZING ON DRIP LOSS, FLAVOR AND COLOR OF BLANCHED AND UNBLANCHED PLEUROTUS FLORIDA

The above experiments demonstrated that Pleurotus florida is minimally affected by enzymatic browning and consequently not likely to require either blanching or sulfiting for color preservation. However, the next step was to ascertain whether the mushrooms could be successfully frozen and stored in an unblanched state. Maintenance of good flavor in frozen storage was, naturally, the ultimate goal. Another objective was to determine whether the rate of freezing would affect drip loss and hence texture. Two freezing regimes were used to study the effect of rapid and slow freezing rates on the amount of drip loss.

5.3.1 Methodology

Pleurotus florida caps (350 g sample size) were frozen in the following manner:

a. Unblanched mushrooms; individually quick frozen in a walk-in freezer at -30°C for 2 hours (air blast),

b. Unblanched mushrooms; slow frozen in bulk in a small cabinet freezer at -18°C for 8 hours (still air),

c. Blanched mushrooms (5 min., steam); also slow frozen in bulk at -18°C as above.

Blanched mushrooms could not be individually quick frozen because of the difficulty in separating the wet mushrooms into individual pieces. All frozen materials were packaged in plastic bags with twist tie closures and stored at -30°C for 4 months.

5.3.1.1 Measurement of Drip Loss After Thawing

Drip loss was measured by placing 100 g of each of the above materials in large funnels over 100 ml graduated cylinders. These were left to thaw at room temperature (23°C) for 2 hours at which time the amount of the drip was determined.

5.3.1.2 Color Evaluation

Three replicates (25 g sample size) of the above materials were evaluated before and after thawing for color (L values; Hunter Colorimeter). These values were compared to control measurements made on blanched and unblanched

Pleurotus florida bulk frozen 24 hours at -30°C to ascertain any possible effects of the frozen storage period.

5.3.1.3 Flavor Evaluation

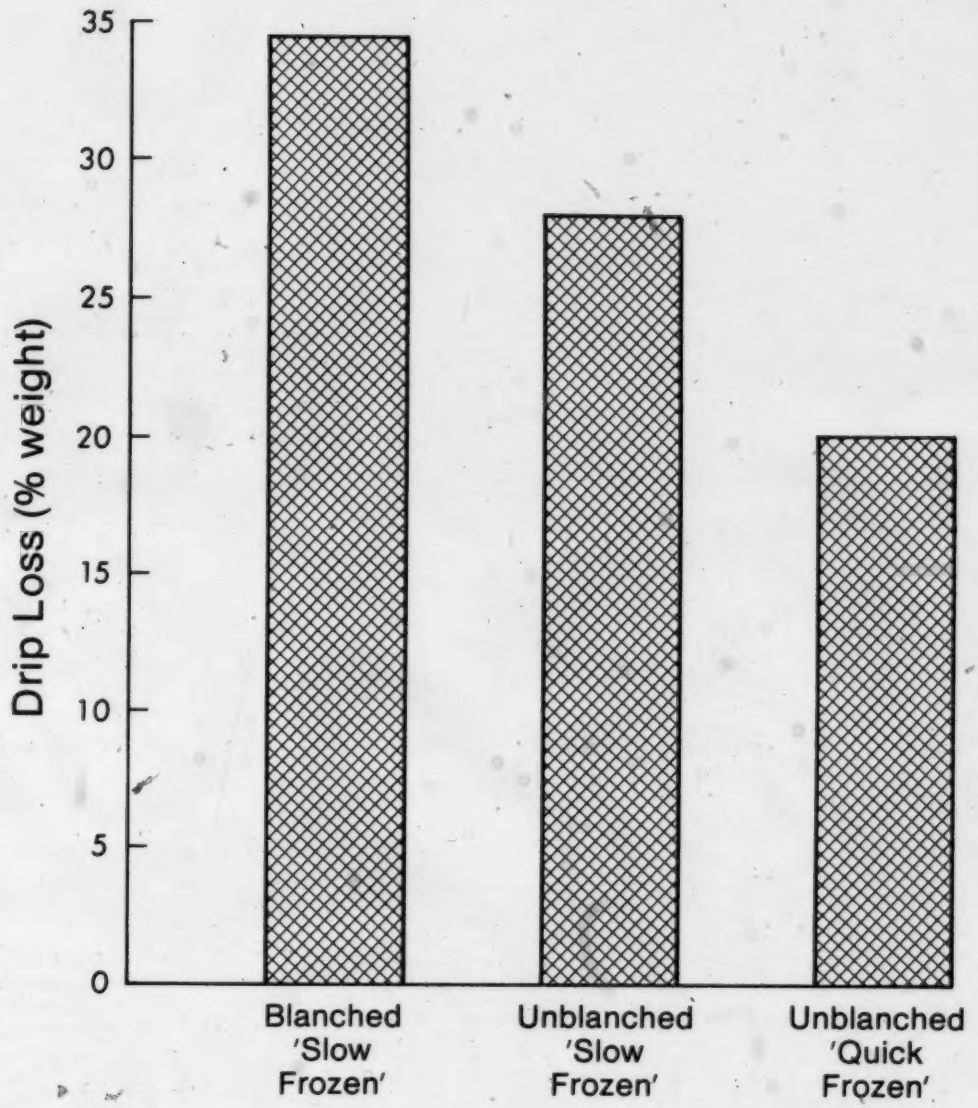
Samples (150 g) of the above materials were thawed at room temperature, sauteed in 10 g of butter for 3 minutes and submitted to taste panel evaluation for flavor assessment (Score Sheet #1, Appendix B). Panelists were asked to rate flavor on a nine point hedonic scale (very good to very poor). Mean scores were calculated and conclusions drawn without the use of statistical analysis.

5.3.2 Results and Discussion

Results for the drip loss experiment are shown in Figure 3. The slow frozen blanched mushrooms showed the greatest degree of drip after thawing, followed by the slow frozen unblanched and finally the quick frozen unblanched. Gormley and O'Riordain (1976) reported drip loss values for Pleurotus florida after freezing and thawing of 16% for unblanched samples and 40% for samples blanched at 100°C for 0.5 minutes. Results in Figure 3 are in good agreement with these values.

According to Fennema (1966) individual quick freezing is the preferred method on the basis of reduced drip after thawing. This is presumably because less intercellular ice is formed and damage to the cells is consequently not as great. Ciobanu et al. (1976) have stated that high freezing

Figure 3 Effect of blanching and freezing rate on drip loss of *Pleurotus florida* after thawing.



rates are especially recommended for mushrooms because of a reduction in the amount of drip. Blanching, which denatures cytoplasmic and membrane proteins, causes an outward diffusion of water. When coupled with the effect of cellular damage due to freezing, it is not difficult to understand why drip is further increased.

Table 11 presents the results obtained for lightness of the materials before and after thawing. In general, the stored blanched mushrooms compared favorably with the blanched control both before and after thawing. The two stored unblanched materials decreased in lightness after thawing, but were still only about 10 units lower than the unblanched control after thawing. These results provided additional evidence that enzymatic browning presents little or no problem in the freezing preservation of Pleurotus florida.

While the above results for drip loss and color measurements appeared to point favorably in the direction of the unblanched mushrooms, some serious problems arose out of the taste panel flavor evaluations (Table 12). Mean scores of poor to very poor were assigned to the two unblanched materials while the blanched one was rated from fair to good. All of the panelists indicated that the former had a strong off-flavor (described as "strawy"). Similar observations on off-flavor development in unblanched Oyster Mushrooms during frozen storage were also made by Gormley and O'Riordain (1976), but no reason for the effect was

Table 11 Lightness of blanched and unblanched Pleurotus florida after four months frozen storage at -30°C.

Sample	Before Thawing (L value)*	After Thawing (L value)*
Unblanched[1]	59.9	46.7
Unblanched[2]	55.9	44.2
Blanched[2]	36.6	34.9
Control (unblanched)[3]	60.2	54.4
Control (blanched)[3]	39.9	38.5

[1] Individually quick frozen (-30°C); stored 4 mo. at -30°C
[2] Slow frozen in bulk (-18°C); stored 4 mo. at -30°C
[3] Frozen in bulk (-30°C); not stored

* Hunter Colorimeter L values

Table 12 Mean taste panel scores for flavor of blanched and unblanched Pleurotus florida after four months frozen storage at -30°C.

Sample	Mean Flavor Score
Unblanched[1]	1.3
Unblanched[2]	1.5
Blanched[2]	7.0

[1] Individually quick frozen (-30°C)
[2] Slow frozen in bulk (-18°C)

advanced.

5.4 THE ROLE OF LIPIDS IN THE DEVELOPMENT OF OFF-FLAVOR DURING FROZEN STORAGE OF UNBLANCHED PLEUROTUS FLORIDA

Development of off-flavors in unblanched or underblanched frozen vegetables has been reported by a number of authors and is believed to be primarily due to the action of lipolytic enzymes (Lee and Wagenknecht, 1951; Lee and Mattick, 1961; Heid and Joslyn, 1967; Ciobanu et al., 1976; Gould, 1977). According to Bennion (1972) and Merritt (1976), either peroxide numbers or thiobarbituric acid (TBA) values can be used to follow the development of off-flavors due to lipid rancidity. In this section, these tests were evaluated for such a purpose and an experiment to monitor the role of the lipid in development of off-flavor during frozen storage of unblanched Pleurotus florida was conducted.

5.4.1 Relationship of Peroxide Numbers and Sensory Evaluation Scores

5.4.1.1 Methodology

The method of Merritt (1976) for TBA determinations was attempted but was found to be unsuitable as all samples produced turbid solutions which could not be properly read by the required spectrophotometric method. Peroxide number tests worked well and preliminary trials were performed on the following materials:

a. Unblanched Pleurotus florida, bulk frozen (-30°C) and stored 9 months at -30°C

b. Blanched (steam; 3 min.) Pleurotus florida, bulk frozen (-30°C) and stored 9 months at -30°C

c. Fresh Pleurotus florida

Lyophilized powders of these materials were prepared and extracted in an all glass Soxhlet apparatus with pure peroxide free anhydrous ethyl ether. The solvent was removed by roto-evaporation and the lipid extracts dried in a vacuum desiccator overnight; triplicate analyses were performed the next day. The micromodified peroxide number method of Lee and Wagenknecht (1951) was employed.

The results of these preliminary trials suggested that lipid rancidity may indeed be responsible for the development of off-flavors. Peroxide numbers of 62.6, 0.0, and 3.0 were found for the unblanched, blanched and fresh materials respectively. On the basis of these observations, this test was utilized for a closer examination of the off-flavor development during frozen storage of the unblanched material.

From a single harvest of Pleurotus florida, successive taste panel flavor evaluations and peroxide number determinations were made. Eight samples (250 g) of the unblanched mushrooms were frozen in bulk and stored at -30°C for 1, 2, 4, and 8 weeks. Four were used for taste panel assessments of flavor; these tests involved hedonic scale evaluations of fresh Pleurotus florida and a thawed frozen

material stored for the appropriate time (Score Sheet #2, Appendix B). A control taste panel evaluation was also made using freshly harvested mushrooms. Both fresh and thawed frozen mushrooms were cooked in 15 grams of butter for 3 minutes before serving. Mean taste panel scores were calculated from the results of these tests and recorded. The remaining four samples were lyophilized at the specified storage time and used for peroxide number tests by the above method.

5.4.1.2 Results and Discussion

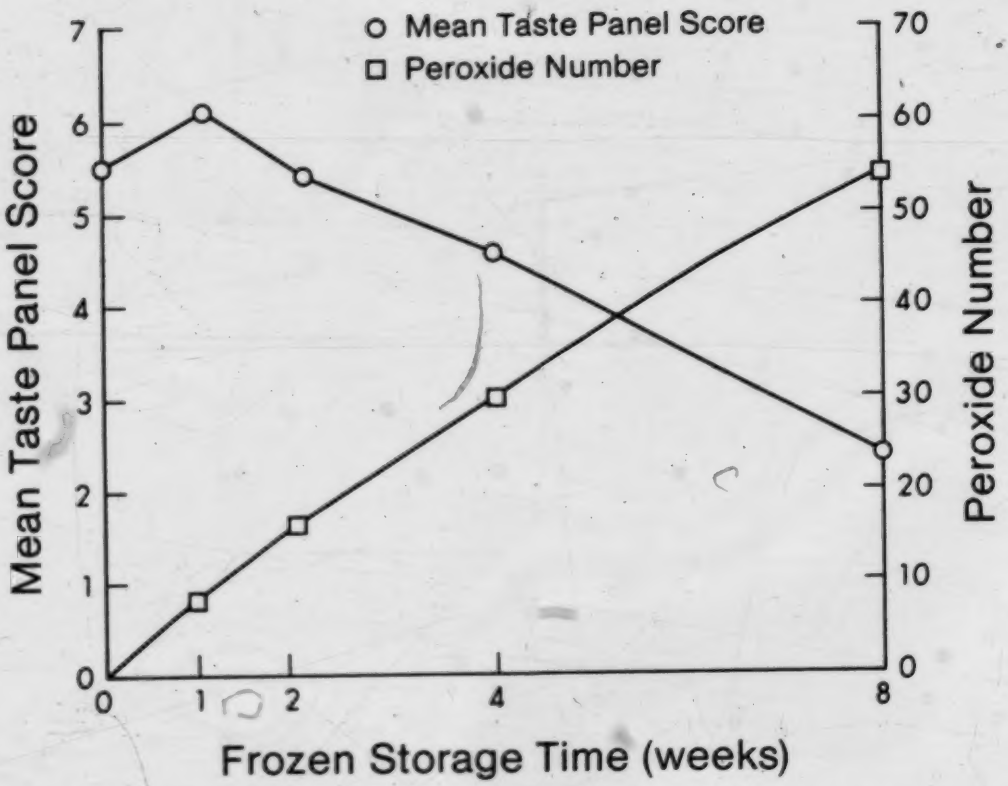
Figure 4 illustrates the correlation between mean taste panel scores and peroxide numbers for the storage period examined. The trend was clear; as peroxide number increased, taste panel scores for the unblanched material decreased. After eight weeks frozen storage, a mean taste panel score of 2.4 (fair to poor) was assigned. Peroxide numbers increased over time until a maximum of 55.20 was obtained at the completion of the storage experiment.

5.4.2 Effect of Frozen Storage on Chemical Changes to the Lipid

5.4.2.1 Methodology

Lipid extracts from the following fresh and stored materials were obtained by the chloroform/methanol extraction method of Bowyer and King (1977):

Figure 4 Relationship of peroxide numbers and mean taste panel flavor scores for unblanched frozen Pleurotus florida.



Extracts 1

a. lyophilized sample prepared from fresh Pleurotus florida,

b. lyophilized sample prepared from unblanched Pleurotus florida (same harvest as above) frozen and stored (-30°C) for 2 months.

Extracts 2

a. fresh Pleurotus florida,

b. unblanched Pleurotus florida frozen and stored for 12 months at -30°C.

Extracts 1 were used to identify the major lipid fractions and their oxidation products. The second set of extracts were used to observe changes in the phospholipid fraction with prolonged storage. All of the extracts were blown dry with nitrogen gas, redissolved in a small amount of benzene and stored at -15°C for use as required.

In identifying the individual lipid fractions and analyzing the fatty acid methyl esters (FAMEs) of Extracts 1, the thin layer chromatography (TLC) solvent system (diethylether/benzene/ethanol/acetic acid) and method of Freeman and West (1966) was employed; plates were spotted with approximately 2000 micrograms of the extracts (2 plates per extract). Individual lipid fractions were located and identified on one set of the TLC plates by their Rf values after development with Rotamine 6G (0.05%). Phospholipid and

neutral lipid fractions were carefully scraped off the set of undeveloped plates, eluted and esterified with boron trifluoride (14% w/w); the FAMEs thus obtained were dissolved in carbon disulphide and two microliters injected into a Bendix Gas Chromatograph 2500 (F.I.D. detector) equipped with an automatic integrator (Spectra-Physics Autolab Minigrator). An 8' glass column packed with Silar 5CP (10%) on Chromosorb W was used for separation of the lipids into their component fatty acids. Inlet temperature was 200°C, column temperature 230°C and detector temperature 250°C. Gas flow rates were: hydrogen - 25 ml/minute, air - 250 ml/minute and nitrogen - 25 ml/minute.

Two TLC plates were quantitatively spotted with 200 micrograms of both of Extracts 2 and chromatographed using the same solvent system as above. To one plate, lipid standards (monoglyceride, diglyceride and a free fatty acid mixture) were applied. Phospholipid fractions were scraped off the second plate and analyzed for phosphorus according to the procedure of Bowyer and King (1977); % phosphorus X 25 was used to convert to % total phospholipid.

5.4.2.2 Results and Discussion

Following TLC separation of Extracts 1, it was apparent that phospholipid comprised a large proportion of the lipid present. The remainder consisted of neutral lipids and sterols.

Results from the GLC fatty acid determinations on the

phospholipid and neutral lipid fractions of Extracts 1 are shown in Figures 5 and 6. In both cases, the lower proportion of unsaturated fatty acids (i.e. 18:1 and 18:2) in the stored sample provided definite evidence that oxidation had taken place during frozen storage.

Plate 10 presents a photograph of the TLC chromatogram where known lipid standards were used to identify some of the spots produced by the stored sample (Extract 2). A very notable observation was the appearance of free fatty acids, most likely due to the breakdown of the phospholipid. This effect is presumed to have arisen from the action of phospholipases. A weaker phospholipid band was also observed for this sample; when checked by chemical analysis, an enormous decrease was found to have occurred as a result of the frozen storage. (The fresh lipid sample had 70% phospholipid while the stored one contained only 20%.)

While other causes of off-flavor development cannot be discounted, these results have demonstrated a change in the composition of the lipid which appears to lead to off-flavors. These off-flavors probably arise as a result of both the presence of free fatty acids and other fatty acid oxidation products. Autoxidation is presumed not to have been a primary cause since no deleterious flavor effects were observed for blanched samples of Pleurotus florida stored for as long as 12 months at -30°C.

Lee and Mattick (1961) reported marked increases in free fatty acids and decreases in fatty acid unsaturation in

Figure 5 Comparison of saturated and unsaturated fatty acid methyl esters (FAMEs) from the neutral lipid fraction of fresh Pleurotus florida and frozen unblanched Pleurotus florida stored for 2 months at -30°C.

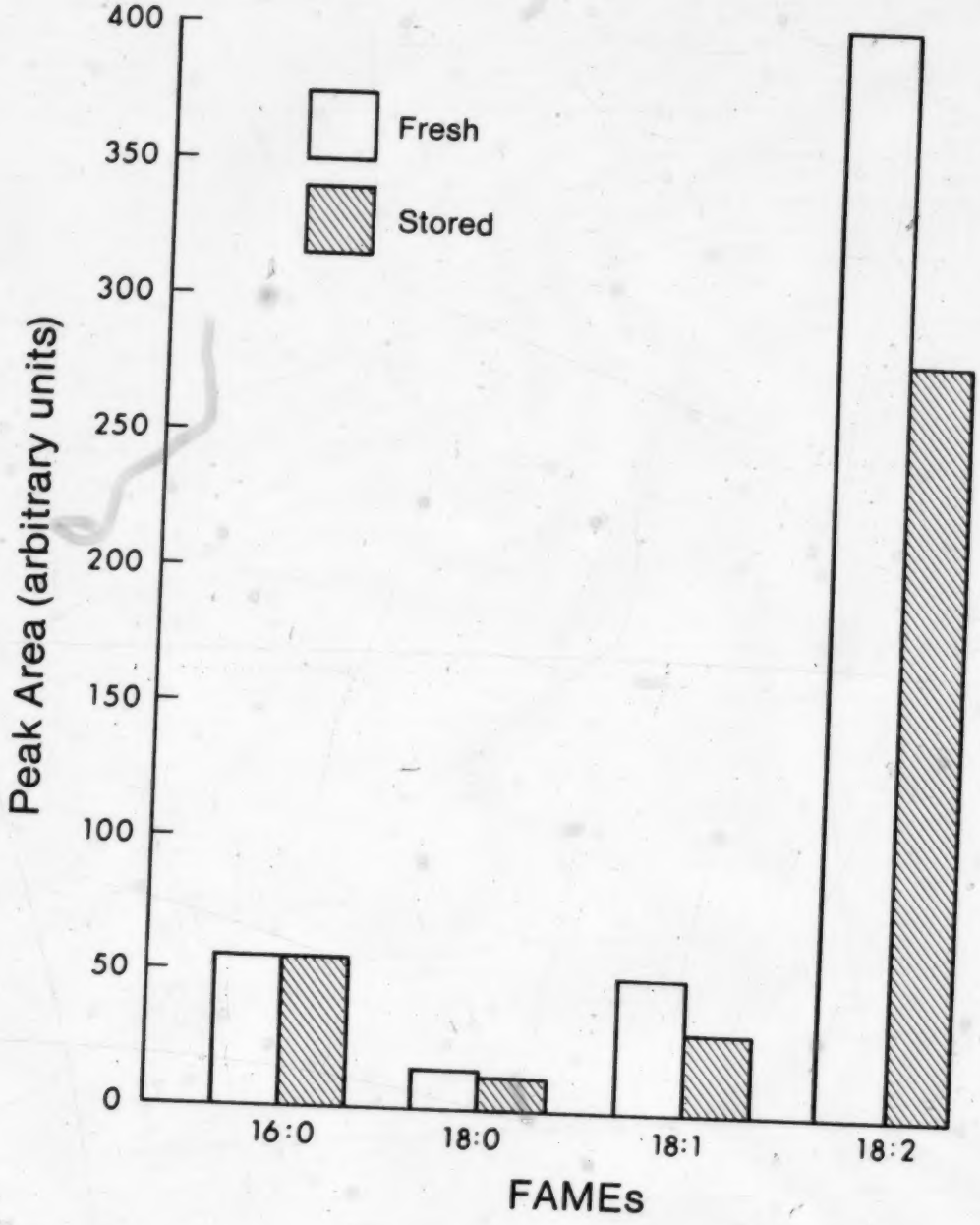


Figure 6 Comparison of saturated and unsaturated fatty acid methyl esters (FAMEs) from the phospholipid fraction of fresh Pleurotus florida and frozen unblanched Pleurotus florida stored for 2 months at -30°C.

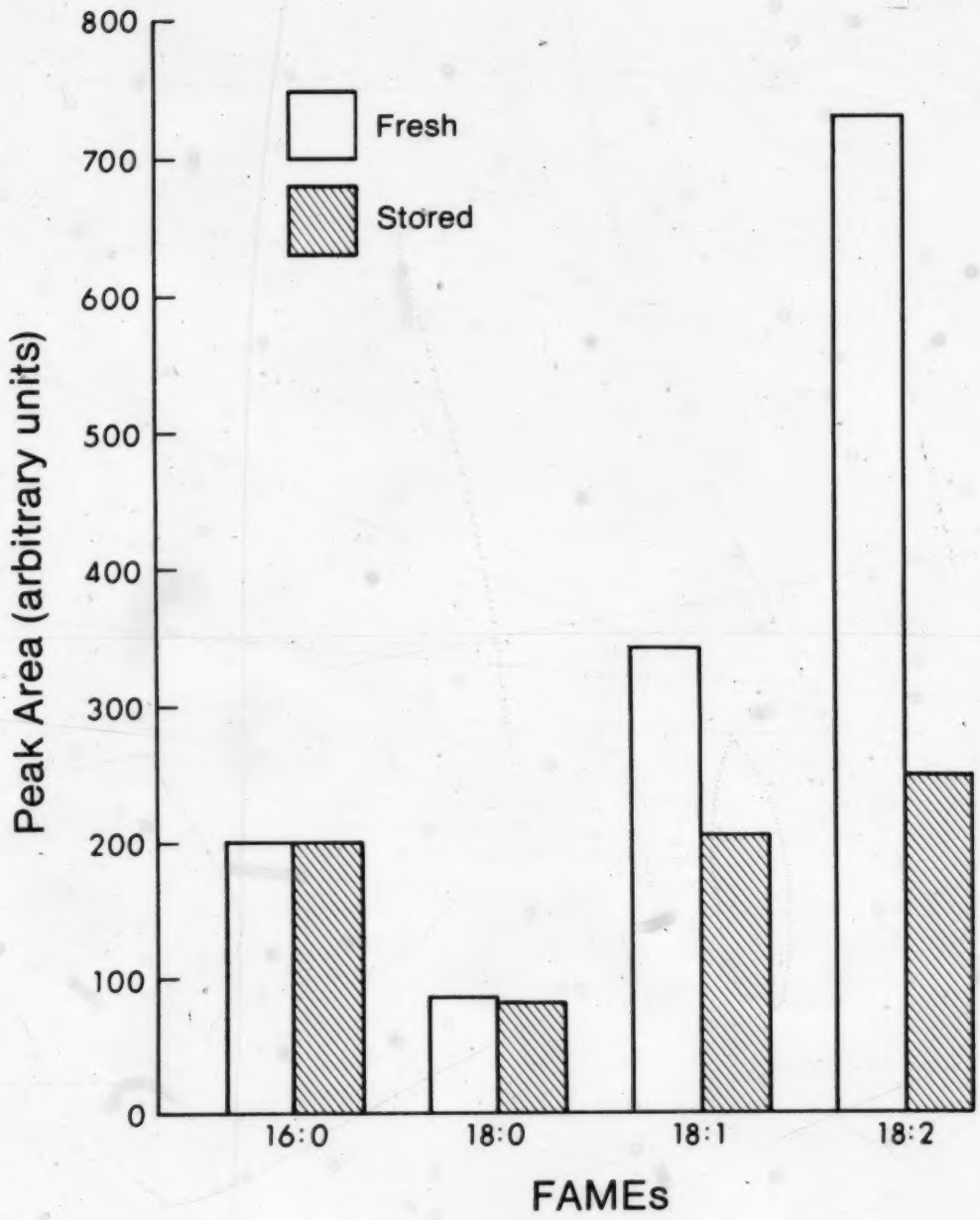
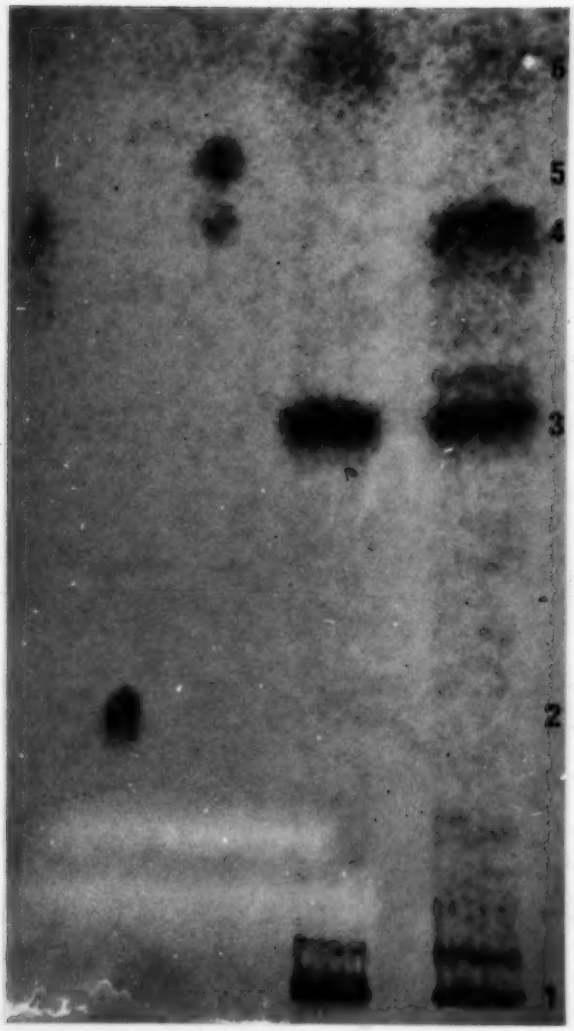


Plate 10 TLC chromatogram of lipid extracts from fresh and stored frozen unblanched Pleurotus florida fruit bodies.



1=Phospholipid
2=Monoglyceride
3=Sterols
4=Free Fatty Acids
5=Diglyceride
6=Triglyceride

STANDARDS SAMPLES
F S

F = Lipid extract from fresh tissue
S = Lipid extract from stored (12 mo. at -30°) unblanched tissue

unblanched peas stored for 1 year at -18°C when compared to enzyme-inactivated samples. They attributed both of these changes to enzymatic causes and related the overall effect to the development of off-flavor. This evidence tends to support the above conclusion. According to Ciobanu et al. (1976), enzymic activity can actually be higher in some frozen tissues than in the fresh produce because the cellular damage caused by freezing brings enzymes into contact with their substrates. Blanching is recommended as the simplest means to overcome this problem.

5.5 TASTE PANEL EVALUATIONS OF FRESHLY FROZEN BLANCHED AND UNBLANCHED PLEUROTUS FLORIDA CAPS

In the above experiments it has been demonstrated that frozen storage of unblanched Pleurotus florida was not successful. However, it was of interest to know whether blanching per se would affect the product quality of the frozen product. For this reason, freshly blanched and unblanched frozen Pleurotus florida were compared to a fresh reference material by taste panel analysis. The questions of interest were:

a. What are the effects of the two methods on tenderness?

b. Is flavor intensity affected by either method?

5.5.1 Methodology

Samples of blanched (steam; 3 min.) and unblanched Pleurotus florida were bulk frozen at -30°C for 24 hours and

cooked without thawing. Fresh mushrooms from the same harvest (stored 24 hours at refrigeration temperature) were used as the reference material. All materials were sauteed in a small amount of butter; fresh for 3 minutes, frozen for 5 minutes. Panelists were asked to evaluate the two test materials by comparing their flavor and texture to that of the reference (Score Sheet 3, Appendix B). Mean scores were calculated and tested for difference against the reference using a t-test. A score of 4 signifies that no difference between the test material and the reference was found.

5.5.2 Results and Discussion

Mean scores and their statistical significance are given in Table 13. It was found that the blanched mushrooms had significantly less flavor ($p \leq 0.05$) and were significantly less tender ($p \leq 0.05$) than the reference. No significant difference was determined in the flavor and texture of the unblanched frozen mushrooms and the reference. It should not be assumed from this data, however, that one method is better than the other. All that has been shown is that slight to moderate decreases in flavor and increases in firmness are likely to occur when the mushrooms are blanched before freezing. Further testing would have to be performed to demonstrate whether these changes are important. Should it be found that blanching is not desirable though, it may be possible to consider the use of other mechanisms for the prevention of enzyme catalyzed

Table 13 Summary of results (mean scores) from the sensory evaluation of freshly frozen blanched and unblanched Pleurotus florida as compared to a fresh reference.

Sample	Flavor	Tenderness
Blanched	4.86*	5.14*
Unblanched	3.86	4.43

*significantly different from reference at 5% level (the reference was arbitrarily assigned a score of 4)

Definition of scores:

1 = considerably more flavor (tender) than the reference
2 = moderately more
3 = slightly more
4 = equal in flavor (tenderness) to the reference
5 = slightly less
6 = moderately less
7 = considerably less flavor (tender) than the reference

lipid changes in the unblanched mushrooms during frozen storage. Approaches such as the incorporation of antioxidants, the exclusion of air from the package and/or the use of chemical inhibitors could be considered.

5.6 REMARKS

Enzymatic browning has been shown not to be a problem in the freezing preservation of *Pleurotus florida* because of low activities of the enzyme PPO and its substrates in the tissue. However, off-flavor development was extensive in unblanched frozen samples stored for as little as two months. Various tests were performed to examine the cause of this problem; changes in the lipid fraction, most likely caused by enzymes, are believed to be primarily responsible.

When mushrooms were blanched prior to freezing, drip loss was greater than that for unblanched frozen samples. Sensory analysis revealed that these mushrooms were less tender and that their flavor intensity was also decreased relative to that of a fresh control. Increasing the freezing rate might be a mechanism for minimizing the effect of textural damage. The freezing preservation of *Pleurotus florida* appears to be fairly difficult with much further work still needed.

6. CANNING EXPERIMENTS

6.1 INTRODUCTION

As mentioned previously, the most widely used preservation technique for mushrooms (*Agaricus bisporus*) in North America is canning. Therefore, preliminary investigations into the suitability of *Pleurotus florida* for canning were conducted.

The main question to be considered was how canning would affect flavor, texture and appearance characteristics of the mushroom. To this end, brine canned products were prepared from the cap and stem portions and subjected to sensory evaluation. The suitability of two other canning mediums was also examined. Finally, sensory characteristics of cream soups, made from either the cap or stem portions of *Pleurotus florida*, were compared to one made with *Agaricus bisporus*.

6.2 METHODOLOGY

Mushrooms from first flush crops of *Pleurotus florida* were separated into cap and stem portions and stored 24 hours at refrigeration temperature to reduce weight loss during blanching and thermal processing. Next, they were blanched in steam for 3 minutes and put into tin cans (size 306 X 411) for hot filling with the customarily used 2% (w/v) salt brine. Sealed cans were then processed in a pilot plant retort (Reid Boiler Works, Bellingham, Wash., U.S.).

Thermocouple measurements were made intially to determine the appropriate total processing time. With a retort load of six cans or less, assuming a required Fo value of 3.5 minutes, this was found to be 12 minutes at 121°C. At the completion of the retorting step, all cans were cooled and stored in a walk-in cooler for use as required in sensory evaluations. Maximum storage time was 1 week.

The first taste panel evaluation was a comparison of fresh caps and fresh stems of Pleurotus florida. This was done to determine the degree of difference between the sensory characteristics of the two portions of the mushrooms. Following that, three taste panel analyses of brine canned and fresh Pleurotus florida were performed:

a. evaluation of canned stems and canned caps,

b. evaluation of fresh caps and canned caps,

c. evaluation of fresh stems and canned stems.

In each case, approximately 150 grams of the fresh or drained canned materials were sauteed in butter (10 g) for 3 minutes. Small containers of these (about 15 g) were then presented to the panelists in a randomly coded fashion. Panelists were asked to rate flavor, texture, appearance and overall characteristics on a 6 point hedonic rating scale of very poor to very good (Score Sheet #4, Appendix B). Results were analyzed by ANOVA and the Duncan's New Multiple Range Test.

Blanched caps of Pleurotus florida were also canned in butter sauce (1 part flour, 3 parts butter, 9 parts water)

and cream sauce (1 part flour, 2 parts butter, 15 parts whole milk) and compared to a brine canned sample prepared at the same time. Salt was added as necessary to assure comparable levels in the three products. All cans were hot filled and processed as above. The following day, the products were reheated and submitted to taste panel evaluation fcr flavor, texture, appearance and overall acceptability ratings using a nine point hedonic scale of very good to very poor (Score Sheet #5, Appendix B). Results were again analyzed by ANOVA and Duncan's tests.

Cream soups made from Pleurotus florida were prepared according to the formula and procedure outlined below:

150 g of mushrooms
28.5 g butter
13.5 g flour
1.0 g salt
500 ml weak chicken broth
150 ml cereal cream

The mushrooms were sauteed in butter for about 3 minutes; salt and flour were blended in and chicken broth added slowly with stirring. The mixture was allowed to simmer gently for about 2 minutes until slightly thickened. Cream was added before blending in a Waring blender for approximately 3 minutes.

A batch was also prepared with fresh Agaricus bisporus (150 g) in the same way. The hot soups were poured into cans and processed 12 minutes at 121°C. Taste panel analysis entailed scoring of flavor and consistency (Score Sheet #6, Appendix B). Appearance evaluation was felt to be of little importance since color can be easily modified as desired. Consequently, coded samples of each hot soup were presented to the panel under red light to mask the effect of appearance. Results were analyzed as described above.

The degree of shrinkage from thermal processing was measured so that a comparison could be made with values determined for Agaricus bisporus. One hundred grams of caps and stems of Pleurotus florida (stored 24 hours post harvest) and slices of Agaricus bisporus were used to prepare brine canned products. Triplicate samples of each material (100 g) were blanched 3 minutes in steam and weighed to determine the weight loss contributed by blanching. After retorting (12 minutes at 121°C) drained product weights were determined according to the method of Luh and Woodroof (1975).

6.3 RESULTS AND DISCUSSION

6.3.1 Sensory Analysis of Fresh Caps and Stems of Pleurotus florida

Results from the statistical analysis of the sensory evaluation of fresh caps and fresh stems of Pleurotus florida are given in Table 14. A highly significant

difference in texture ($p \leq 0.01$) was found with caps being rated more acceptable than stems. No other differences were statistically significant. Comments on the texture of the stems such as "woody", "rubbery" and "chewy" were noted. Their high fiber content (see Table 6) is most likely responsible for this effect. It seemed that if the texture of the stems could be softened by heat processing, then canning might be a viable way to utilize this portion of the mushroom.

6.3.2 Sensory Analysis of Brine Canned Caps and Stems of *Pleurotus florida*

Statistical analysis of the results obtained for the taste panel comparison of brine canned caps and brine canned stems of *Pleurotus florida* is shown in Table 15. Differences in both texture and overall scores of the two were highly significant ($p \leq 0.01$) with stems being rated significantly better than caps. It would appear that the improvement in texture of the canned stems substantially influenced the scores assigned by the panelists for their overall quality.

The next step was to compare brine canned samples of each of caps and stems to freshly cooked counterparts. This was done to answer the question of how acceptable the canned products are on a relative basis. Results for the statistical analyses are given in Tables 16 and 17. The comparison of fresh and canned caps (Table 16) showed that the fresh caps were rated significantly better in terms of

Table 14 Summary of results (mean scores) from the sensory evaluation of fresh caps and fresh stems of Pleurotus florida.

	Attributes			
	Flavor	Texture	Appearance	Overall
Caps	4.6	5.4**	4.4	4.8
Stems	4.8	3.8	4.6	4.5

* significantly different at 5% level
** significantly different at 1% level

Table 15 Summary of results (mean scores) from the sensory evaluation of canned caps and canned stems of Pleurotus florida.

	Attributes			
	Flavor	Texture	Appearance	Overall
Caps	3.6	4.4	4.5	3.8
Stems	4.6	5.4**	4.8	5.0**

* significantly different at 5% level
** significantly different at 1% level

Table 16 Summary of results (mean scores) from the sensory evaluation of fresh and canned caps of Pleurotus florida.

	Attributes			
	Flavor	Texture	Appearance	Overall
Fresh caps	5.1**	4.8**	4.5*	4.8**
Canned caps	3.4	4.1	4.0	4.0

* significantly different at 5% level
** significantly different at 1% level

Table 17 Summary of results (mean scores) from the sensory evaluation of fresh and canned stems of Pleurotus florida.

	Attributes			
	Flavor	Texture	Appearance	Overall
Fresh stems	3.8	4.6	4.2	4.0
Canned stems	4.6	5.6*	4.8	5.0**

* significantly different at 5% level
** significantly different at 1% level

flavor, texture, overall acceptability ($p \leq 0.01$ in each case) and appearance ($p \leq 0.05$). These results demonstrated that brine canned caps are not nearly as acceptable as the fresh mushrooms. Comments indicated that the flavor of the brine canned mushrooms was "off" or "sour" and that they were too "mushy" or "sloppy" looking.

Table 17 shows the results from the comparison of fresh and canned stems. Differences between the two were again found to be significant ($p \leq 0.01$), but here the trend was reversed from that found for the caps. In this case, canned stems were rated significantly better than fresh stems in terms of texture ($p \leq 0.05$) and overall scores ($p \leq 0.01$). It appears that canning substantially improved the texture of the stems and that consequently they were more acceptable overall than the fresh ones. The solubilization of pectic substances in the cell walls of vegetable tissues is known to be largely responsible for the effect of texture softening caused by thermal processing. However, the presence or absence of pectic substances in fungi has not been conclusively demonstrated (Aronson, 1965).

6.3.3 Sensory Analysis of Cream and Butter Sauce Canned *Pleurotus florida*

Because of the low scores that were given to brine canned caps of *Pleurotus florida*, it was decided that other canning packs should also be examined. To this end, canned products made with cream and butter sauces were compared to

a brine canned product by sensory analysis. From the statistical analysis of the results (Table 18), it is apparent that either of the two sauces significantly improved the acceptability of Pleurotus florida caps. Flavor of the brine canned product was rated significantly lower ($p \leq 0.01$) as was the overall score ($p \leq 0.01$). Appearance of the cream sauce product was rated significantly higher ($p \leq 0.05$), indicating that the masking effect is of a positive nature. No significant differences in texture were found.

6.3.4 Cream of Mushroom Soups: Comparison of Pleurotus florida and Agaricus bisporus

As a final test on the suitability of Pleurotus florida for canning, cream of mushroom soups made from Pleurotus florida caps and stems and Agaricus bisporus (stems and caps together) were compared. A summary of the results from this test is shown in Table 19.

The mean score for soup made from Pleurotus florida caps was somewhat lower than the other two, but the difference was not significant. The conclusion is that although the flavor of the two mushrooms is different, the results indicate that either mushroom is equally acceptable in soup form. In the case of the consistency scores, it is interesting to note the slightly higher mean score for soup made from Pleurotus florida stems. The greater thickness of this soup (possibly due to the high fiber content of the

Table 18 Summary of results (mean scores) from the sensory evaluation of brine, butter sauce and cream sauce canned Pleurotus florida caps.

	Attributes			
	Flavor	Texture	Appearance	Overall
Brine	4.0**	6.6	5.0	4.6**
Butter sauce	7.4	6.6	5.8	7.0
Cream sauce	7.0	6.8	7.2*	7.2

* significantly different at 5% level
** significantly different at 1% level

Table 19 Summary of results (mean scores) from the sensory evaluation of soups made from Pleurotus florida caps, Pleurotus florida stems and Agaricus bisporus.

	Attributes	
	Flavor	Consistency
Pleurotus florida caps	5.8	6.6
Pleurotus florida stems	7.2	7.1
Agaricus bisporus	7.4	6.4

* significantly different at 5% level
** significantly different at 1% level

stems) may be the desirable characteristic.

6.3.5 Determination of Weight Loss After Brine Canning: Pleurotus florida versus Agaricus bisporus

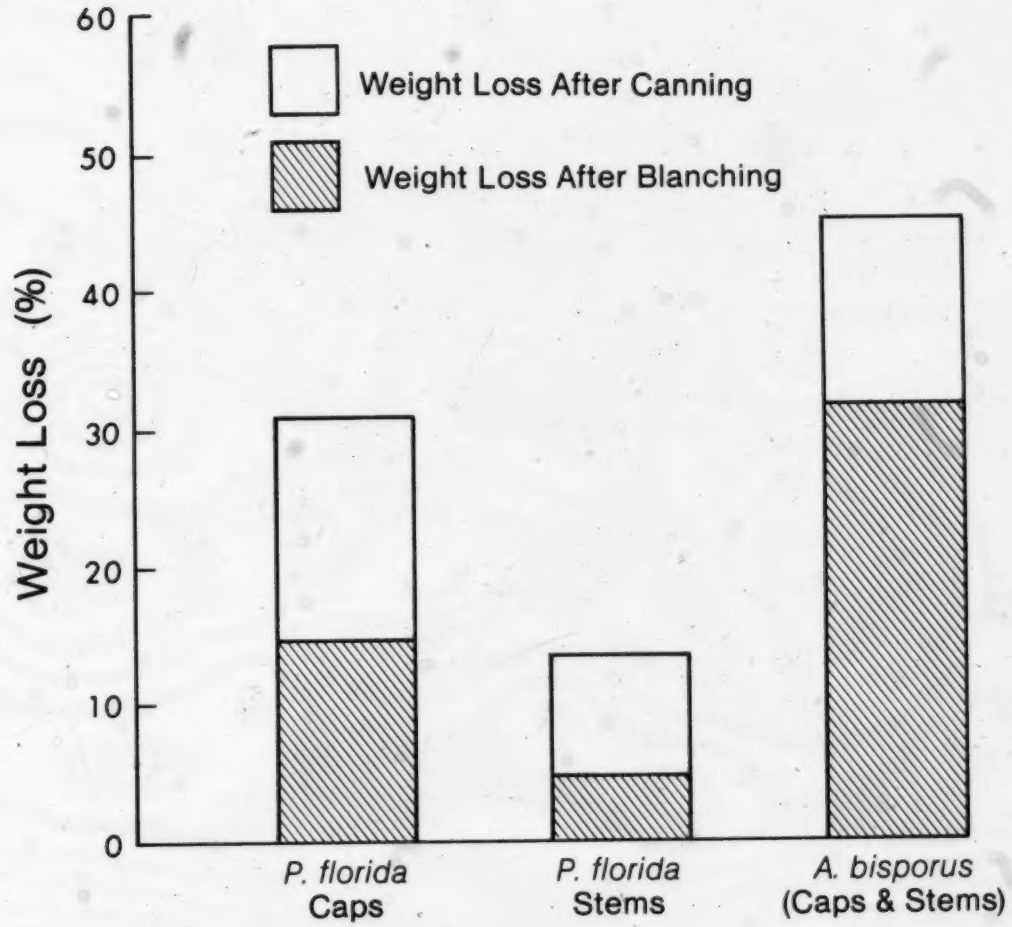
Data for weight loss of Pleurotus florida and Agaricus bisporus after blanching and brine canning treatments are shown in Figure 7. The average total shrinkage for the stem portion of Pleurotus florida was about half that found for the cap portion, and over three times lower than that found for Agaricus bisporus. Determinations for Agaricus bisporus indicated a total shrinkage of about 45% which is only slightly greater than that found by McArdle and Curwen (1962) in similar blanching/canning experiments. Andreotti et al. (1975) found the shrinkage of Pleurotus ostreatus after blanching and canning to be approximately 50% less than would normally be expected for Agaricus bisporus.

6.4 REMARKS

Only one reference to canning experiments with the Oyster Mushroom was located in the course of this project. The authors (Andreotti et al., 1975), claim that Pleurotus ostreatus is very suitable for canning because of its high fiber content. Two canning packs were tested: a) brine, and b) oil and vinegar with garlic, parsley, oregano and salt. No data on sensory analysis were reported.

In the present work, brine canned Pleurotus florida caps have been shown by sensory analysis to be significantly

Figure 7 Weight loss after blanching and brine canning of Pleurotus florida and Agaricus bisporus.



less acceptable than fresh caps. Indications were that both the flavor and texture were detrimentally affected by heat processing. Cream and butter sauces were found to be significantly more acceptable than brine, but further work is needed before conclusive statements on their acceptability can be made. Cream soups prepared from Pleurotus florida scored favorably when compared to one formulated with Agaricus bisporus; this application may warrant further consideration. Where stems were concerned, canning in brine substantially improved their acceptability rating over the fresh ones. All conclusions in this section of the work, however, must be viewed as tentative because of the size and representativeness of the panel and also because results are based on only one replicate per test.

A notable characteristic of Pleurotus florida was its lower level of weight shrinkage in the combined treatment of blanching and brine canning. Average shrinkage values for the caps and stems respectively were 30% and 70% lower than levels determined for Agaricus bisporus. This information may be useful if a suitable canned product can be developed from the Oyster Mushroom.

7. DRYING EXPERIMENTS

7.1 INTRODUCTION

Although dehydrated mushrooms are not an important commercial product in North America, certain amounts are produced for various uses. Examples of their application are as ingredients in dehydrated soups and sauce mixes (eg. pizza sauce, spaghetti sauce, gravy base etc.). Although limited emphasis was placed on the investigation of the potential of *Pleurotus florida* for drying in the present work, some tests were performed because of its seemingly inherent advantages over *Agaricus bisporus* for such a use.

In the first place, *Pleurotus florida* has been shown to exhibit a much lower susceptibility to enzymatic browning (see section 5.2). Hence, the use of blanching and/or chemical preservatives is not likely to be necessary for color preservation during drying or upon rehydration. Secondly, it should lend itself well to rapid and easy removal of moisture because of a large surface area and thin cross section. Last, but not least, fresh *Pleurotus florida* contains high concentrations of the flavor components 1-Octen-3-ol and 5'-GMP (Dijkstra, 1976); these levels may serve to enhance their flavor after dehydration relative to that of other varieties.

7.2 DRYING METHODS AND PRODUCT CHARACTERISTICS

7.2.1 Sun Drying

On several occassions, unblanched Pleurotus florida caps were dried on fine mesh screens in direct sunlight (average drying time was about 6 to 8 hours). Air temperature was normally 20 - 25°C and relative humidity ranged from 30% to 50%. Moisture content of the dried product was measured as described in section 3.4 and averaged about 5 - 7%. Color of the mushrooms was dark to light tan at the completion of the drying operation.

In a similar manner, one batch of blanched Pleurotus florida (steam; 3 min.) was also prepared. It was found, however, that drying took a much longer time and that, in addition, final appearance was much less acceptable. The longer drying time was largely due to the fact that blanching severely compacted the mushrooms. Under these conditions, moisture removal was extremely difficult; when dry, the mushrooms were very dark in color and had a semi-translucent appearance.

7.2.2 Hot Air Drying

Cabinet drying at 65°C for 3 hours was also tried in a few instances using unblanched Pleurotus florida caps and was shown to produce a product very similar to the sun dried one. The slightly greater level of brownish-yellow color development which was observed can probably be attributed to non-enzymatic browning. The method of two stage drying

(first lower and then higher temperature), as used by Komanowsky et al. (1970) for Agaricus bisporus, might minimize this effect.

7.2.3 Freeze Drying

Unblanched Pleurotus florida caps were frozen at -40°C and dried in an RePP freeze drier (Virtis Co. Inc., Gardiner, N.Y.) for 24 hours (shelf temperature = 35°C, fluid temperature = 140°C and condensor temperature = -60°C). The product was beautifully white in color, but lacked the rigidity of the sun dried mushrooms. Their very fragile nature caused them to be easily broken and great care had to be taken in packaging and storage. After rehydration they became very limp and did not retain their shape well. Moisture content after drying was approximately 2 - 3%.

7.2.4 Other Drying Methods

Two other drying methods, namely drum drying and fluidized bed drying, were attempted.

In the case of drum drying, it was found that 3 parts of blanched mushrooms could be comminuted in a Waring blender with 1 part water to make a slurry that was easily dried. Unblanched mushrooms, on the other hand, had to be combined in a ratio of 2 parts mushrooms to 1 part water for blending and could not be successfully dried by this means because of the high water content. In the former case, color

of the resultant product was darker than that of the hot air dried mushrooms. This indicated that non-enzymatic browning was more pronounced, probably because of the high temperature of the drum surface. Still, their color was considerably lighter than that of a similarly prepared drum dried Agaricus bisporus sample which was a dark brownish black. The flavor of the drum dried product was more 'cooked' than that of products dried by the other methods. Nevertheless, it may be a technique worthy of further investigation.

Attempts were made to use fluidized bed drying but to no avail. Even with very finely chopped pieces, the high moisture content prohibited adequate fluidization. After 30 minutes at 65°C with maximum air circulation, moisture content was still 25%. Fluidized bed drying, at least as it was used in these trials, did not appear to be a suitable method for drying Pleurotus florida.

7.3 TASTE PANEL EVALUATIONS OF FLAVOR AND TEXTURE OF FREEZE DRIED AND SUN DRIED PLEUROTUS FLORIDA

7.3.1 Methodology

Samples of unblanched sun dried and freeze dried Pleurotus florida, prepared as described above, were rehydrated in 10 volumes of water at room temperature for 10 minutes. The freeze dried mushrooms were extremely soggy and excess moisture had to be removed by gentle squeezing. Both samples were drained for 5 minutes before being sauteed in a

small amount of butter for 5 minutes. Freshly harvested Pleurotus florida, also cooked in butter for 5 minutes, was used as the reference material. Panelists were asked to compare the flavor and texture of the two dried products to those of the reference using Score Sheet #7, Appendix B. Mean scores were calculated and tested for difference against the reference using a t-test; a score of 5 means that the dried product was rated as being equal to the reference. The panelists were also asked to state their preference for texture.

7.3.2 Results and Discussion

Statistical analysis of the data from the sensory evaluation of sun and freeze dried Pleurotus florida are shown in Table 20. Only the texture of the sun dried material was found to be significantly different from that of the reference; it was rated as being 'much' to 'moderately' less tender. On the average, the panelists found the flavor of both dried products to be comparable or slightly inferior to the reference, although the difference was not statistically significant. With regard to the supplementary question on preference for texture, three panelists stated that they found the reference more acceptable, two preferred the sun dried mushrooms and one the freeze dried mushrooms. This observation was consistent with the previous finding that the panel exhibited no clear cut preference trend for texture. Those who chose the sun

Table 20 Summary of results (mean scores) from the sensory evaluation of sun dried and freeze dried Pleurotus florida as compared to a fresh reference.

Sample	Flavor	Tenderness
Sun dried	5.5	7.7**
Freeze dried	5.2	5.5

**significantly different from reference at 1% level (the reference was arbitrarily assigned a score of 5)

Definition of scores:

1 = Extremely superior flavor (more tender) than the reference
2 = Much superior flavor (more tender)
3 = Moderately superior flavor (more tender)
4 = Slightly superior flavor (more tender)
5 = Equal to the reference in flavor (tenderness)
6 = Slightly inferior flavor (less tender)
7 = Moderately inferior flavor (less tender)
8 = Much inferior flavor (less tender)
9 = Extremely inferior flavor (less tender) than the reference

dried product commented that they liked the 'firmness'. Others, however, found it less acceptable than the reference or the freeze dried one. Two panelists indicated that the flavor of the sun dried product was "more mushroomy".

7.4 EFFECT OF STORAGE ON SUN DRIED PLEUROTUS FLORIDA

7.4.1 Methodology

Blanched (steam; 3 min) and unblanched *Pleurotus florida* were sun dried according to the method described above. Samples of each were placed into sealed polyethylene bags and stored at 25°C for 10 months. One batch of freshly dried unblanched material was used for control measurements.

Rehydration ratio is defined as the weight of water (g) absorbed per unit weight (g) of dried material. These ratios were determined for the control and the two stored materials in the following way. Ten grams of each material were placed into beakers containing 500 ml of water at room temperature. The mushrooms were removed at 30 second intervals, blotted on a single thickness of paper towel for a standard time to remove excess surface moisture and reweighed until no further weight gain was evident; the total rehydration time was recorded. Lightness values of the rehydrated materials were determined in duplicate using the Hunter Colorimeter.

Sensory evaluation, using a triangle test for difference, was conducted to determine if flavor had been affected by storage. One sample of the unblanched dried mushrooms was presented along with two freshly prepared

unblanched sun dried controls; all were rehydrated in water for 5 minutes and sauteed in a small amount of butter for 3 minutes before serving. Panelists were asked to choose the odd sample on the basis of flavor difference using Score Sheet #8 (Appendix B). Results were collected and statistical significance assessed by consulting the appropriate tables (ASTM, 1969).

7.4.2 Results and Discussion

Results for the rehydration ratio and lightness measurements are given in Table 21. The fresh sun dried control was the lightest and had the highest rehydration ratio. The value for the stored unblanched material, however, was only slightly lower and in general compared favorably with the control. The blanched sample, on the other hand, scored extremely low in terms of both rehydration time and rehydration ratio. The color of this sample (dark brown) was not measured in the Hunter Colorimeter as it was so obviously less acceptable. Blanching seems to impart a number of undesirable side effects and its use as a pretreatment for sun drying is not recommended on this basis.

With regard to the taste panel analysis of flavor, only 5 out of the 8 panelists correctly identified the odd sample. This was an insufficient number to establish statistical significance at any level. It is therefore concluded that no deleterious flavor changes occurred in the

Table 21 Rehydration ratio, time and color of blanched and unblanched sun dried Pleurotus florida after storage.

Sample	Rehydration ratio (Water:dry material)	Rehydration time (minutes)	Color (L)*
a	6.1:1	2	45
b	5.8:1	2	43
c	2.5:1	8	-

a = unblanched control; not stored
b = unblanched; stored 10 months at 25°C
c = blanched; stored 10 months at 25°C

* L values on Hunter Colorimeter

unblanched sun dried product over the storage period examined. This finding supports the previously advanced hypothesis that development of fat rancidity is not due to autooxidative processes only. The low water content and water activity of the dried material probably severely limit the extent of enzymatic activity and related off-flavor production.

7.5 REMARKS

It is the author's opinion that sun drying of unblanched *Pleurotus florida* showed considerable promise as a dehydration method. The reasons for this conclusion are due to the following:

a. Results from the sensory evaluation of sun dried mushrooms were generally favorable.

b. Freeze dried *Pleurotus florida* was much too fragile to be packaged by conventional means. In addition, they did not rehydrate in a satisfactory manner. Neither of these problems were apparent for sun dried samples.

c. Sun drying is likely to have distinct advantages over freeze drying from a production standpoint (i.e. it is an easier and cheaper method).

d. Storage tests demonstrated that the sun dried product was stable in terms of the quality parameters measured for periods of up to 10 months.

The use of appropriately designed cabinet or tray drying

methods would no doubt be equally suitable. Sun or hot air drying may well prove to be the most suitable preservation techniques for the Oyster Mushroom.

8. CONCLUSIONS AND RECOMMENDATIONS

8.1 SUMMARY OF RESEARCH FINDINGS

The investigations in the present work were primarily directed at evaluating the processing potential of the Oyster Mushroom, Pleurotus florida. Much of the work was exploratory in nature as a limited amount of research has been devoted to this topic to date.

Although not directly related to the above objective, an additional contribution was the refinement and testing of an easily reproducible cultivation method. This is believed to have been a necessary undertaking since much of the information in the literature is in languages other than English and also because frequently descriptions are not comprehensive or explicit.

Proximate analyses were performed on both the stem and cap portions of Pleurotus florida from first and second flush crops. This aspect, until now, has not been carefully considered as to its effect on the ultimate outcome of the analysis. The results obtained show that this is fundamentally important for proper sampling techniques. As a general trend, it was found that protein content is higher and fiber content lower in caps than in stems. The main differences between first and second flushes were that protein and ash contents decreased and fiber level increased in the second flush mushrooms. Crude protein values for first flush Pleurotus florida caps were found to be very

comparable to those quoted in the literature for Agaricus bisporus (each about 40% on a dry weight basis); others have quoted lower values for the Oyster Mushroom. Another observation was that the traditional method for fat analysis (ether extraction) proved to be inadequate. The presence of high levels of polar lipids prohibited complete recovery unless a mixed solvent system was used.

Blanching experiments on Pleurotus florida showed that steam and water blanching have different effects on the level of shrinkage and total solids content. Water blanching resulted in a lower weight loss but leached about 31% of the solids. The portion of the mushrooms and the flush were also shown to affect the degree of shrinkage. In general, stems were found to be less susceptible than caps to blanching shrinkage, as were second flush materials. Pleurotus florida exhibited a significantly lower level of weight loss after blanching than Agaricus bisporus. With post harvest storage, this difference was further enhanced; weight loss values of 13% and 25% were found for Pleurotus florida and Agaricus bisporus respectively.

Experiments on the freezing preservation of Pleurotus florida demonstrated that blanching is necessary for inactivation of enzymes which cause substantial off-flavor development in frozen storage. Chromatographic studies showed the oxidation of lipid, with accompanying production of free fatty acids, to be extensive in unblanched frozen samples stored for various periods of time; these changes

are thought to be primarily responsible for the development of off-flavor during frozen storage. Taste panel evaluations of Pleurotus florida, blanched prior to freezing, showed that flavor intensity and tenderness were decreased relative to that of a fresh reference material.

With regard to the potential for canning of Pleurotus florida, it was found that brine may not be the most suitable pack. Butter and cream sauces were also evaluated and both resulted in a more acceptable product from a flavor standpoint. Cream sauce showed the greatest potential, largely because of its positive influence on appearance. Fresh stems were found to be unacceptably fibrous but canning markedly improved their status; canning is suggested as a possibly viable means for their utilization. The use of Pleurotus florida in a cream of mushroom soup product compared favorably with one made from Agaricus bisporus. Both the caps and stems of Pleurotus florida lost significantly less weight than Agaricus bisporus in the combined treatment of blanching and thermal preservation.

Dehydration showed excellent prospect as a preservation technique for Pleurotus florida, particularly if simple methods were used (ie. sun or hot air drying). Sensory analysis of sun dried Pleurotus florida showed that the rehydrated product was significantly less tender than fresh Pleurotus florida but that this is not necessarily an undesirable characteristic. No difference in flavor was determined. Blanching appears to be unnecessary for color

preservation of Pleurotus florida because PPO and its substrates occur in low concentration. In fact, it appeared to be undesirable as a pretreatment for sun drying; drying time increased and rehydration ratio and time were poor when blanching was used. A sun dried product made from unblanched Pleurotus florida had good rehydration characteristics with acceptable color and storage life.

To summarize, the main advantages of Pleurotus florida for processing are:

a. easy cultivation
b. minimal levels of enzymatic browning
c. low blanching and canning shrinkage
d. adaptability to simple drying methods
e. potential for some canned products

Freezing preservation poses the biggest problem. While it would be highly desirable to use unblanched Pleurotus florida for a number of reasons, off-flavor development during even short term storage is serious.

8.2 SUGGESTIONS FOR FURTHER WORK

It is recommended that the development of suitable and effective objective testing methods for flavor and texture assessment would be extremely useful. This project relied heavily on the use of subjective measurements; the conclusions drawn from it should probably be substantiated by the the use of appropriate objective measurements. Gas chromatography head space analysis shows promise as a method

for flavor profile work. Texture measurements are more difficult. The unusual shape and wide variations in fruit body sizes make most conventional methods, such as the use of the Instron testing machine, unsuitable. Microscopic studies on the tissue of Pleurotus florida, performed before and after blanching, freezing, canning and/or drying, may clarify or explain the effects of these treatments on cellular changes. They could also serve as an indirect method for texture assesment.

The findings on the problem of fat rancidity in unblanched frozen Pleurotus florida were very intriguing. Additional investigations might include analyses to determine the exact nature and relative activities of the enzymes involved. A thorough assessment and evaluation of preventative treatments other than blanching would probably be a worthy undertaking.

In order to assess the acceptability and preference patterns of processed products made from the Oyster Mushroom, all operations would have to be scaled up and carefully evaluated. Extensive market testing and product development work would be required if interest warranted it.

The aspect of utilization of the spent substrate as an animal feed also deserves emphasis. This feature, if shown to be feasible, would undoubtably make the economics of mushroom production more favorable.

Finally, it may be useful to consider the possibilities for improvement of this mushroom via breeding work. Various

species and strains of the Oyster Mushroom may well have uniquely advantageous characteristics. The development of new varieties more suitable for industrial processing might well be the clue to the marketing breakthrough for this new food material.

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10. APPENDIX A

This appendix contains the results of the analysis of variance tests on the data from the weight loss experiments performed on Pleurotus florida and Agaricus bisporus in the blanching study.

Table A Shrinkage of *Pleurotus florida* (1st and 2nd flushes) by steam and water blanching: stems vs. caps.

Source of variation	d.f.	S.S.	M.S.	F
stems vs. caps	1	443.8	443.8	8.6**
replicates	2	7.0	3.5	0.1
runs	2	291.1	145.6	2.8
treatment&flush (TF)	3	1286.4	428.8	8.4**
replicates X runs	4	11.2	2.8	0.05
replicates X TF	6	20.9	3.5	0.1
runs X TF	6	225.3	37.5	0.7
replicates X runs X TF	12	76.6	6.4	0.1
error	35	1794.9	51.3	
total	71	4157.2	0	

* significant at 5% level

** significant at 1% level

Table B Shrinkage of Pleurotus florida (stems and caps from 1st and 2nd flushes): water vs. steam blanching.

Source of variation	d.f.	S.S.	M.S.	F
water vs. steam	1	1337.5	1337.5	8.6**
replicates	2	6.3	3.2	0.02
runs	2	486.9	243.4	1.6
samples&flush (SF)	3	3146.9	1049.0	6.7**
replicates X runs	4	9.3	2.3	0.2
replicates X SF	6	46.4	7.7	0.7
runs X SF	6	935.5	155.9	13.7**
replicates X runs X SF	12	74.6	6.2	0.5
error	35	398.4	11.4	
total	71	6441.7	0	

* significant at 5% level

** significant at 1% level

Table C Shrinkage of Pleurotus florida (stems and caps) by steam and water blanching: 1st flush vs. 2nd flush.

Source of variation	d.f.	S.S.	M.S.	F
flush vs. flush	1	2289.8	2289.8	49.6**
replicates	2	6.3	3.2	0.06
runs	2	486.9	243.4	5.3**
samples&treatment (ST)	3	1782.4	594.1	12.9**
replicates X runs	4	9.3	2.3	0.05
replicates X ST	6	20.3	3.4	0.07
runs X ST	6	156.8	26.1	0.6
replicates X runs X ST	12	74.8	6.2	0.1
error	35	1615.0	46.1	
total	71	6441.7	0	

* significant at 5% level

** significant at 1% level

Table D Shrinkage of Agaricus bisporus by steam and water blanching: stems vs. caps.

Source of variation	d.f.	S.S.	M.S.	F
stems vs. caps	1	1072.6	1072.6	11.49**
replicates	2	1.2	0.6	0.01
runs	2	76.2	38.1	0.4
treatment (T)	1	22.9	22.9	0.2
replicates X runs	4	44.3	11.1	0.05
replicates X T	2	71.5	35.8	1.6
runs X T	2	186.6	93.3	4.2**
replicates X runs X T	4	60.1	15.0	0.7
error	17	377.2	22.2	
total	35	1912.8	0	

* significant at 5% level

** significant at 1% level

Table E Shrinkage of Agaricus bisporus stems and caps: water vs. steam blanching.

Source of variation	d.f.	S.S.	M.S.	F
water vs. steam	1	22.9	22.9	0.9
replicates	2	1.2	0.6	0.02
runs	2	76.2	38.1	1.4
samples (S)	1	1072.6	1072.6	41.1**
replicates X runs	4	44.3	11.1	0.4
replicates X S	2	76.3	38.2	1.5
runs X S	2	50.2	25.1	1.0
replicates X runs X S	4	124.7	31.2	1.2
error	17	444.2	26.1	
total	35	1912.8	0	

* significant at 5% level

** significant at 1% level

Table F Shrinkage of stems and caps by steam and water blanching: Agaricus bisporus vs. Pleurotus florida (1st flush).

Source of variation	d.f.	S.S.	M.S.	F
A. bisporus vs. P. florida	1	125.4	125.4	4.4*
replicates	2	7.5	3.8	0.1
runs	2	492.1	246.0	8.7**
samples&treatment (ST)	3	1847.2	615.8	21.8**
replicates X runs	4	12.1	3.0	0.1
replicates X ST	6	89.9	15.0	0.5
runs X ST	6	183.6	30.6	1.1
replicates X runs X ST	12	207.1	17.2	0.6
error	35	987.9	28.2	
total	71	3952.9	0	

* significant at 5% level

** significant at 1% level

Table G Shrinkage of stems and caps by steam and water blaching: *Agaricus bisporus* vs. *Pleurotus florida* (2nd flush).

Source of variation	d.f.	S.S.	M.S.	F
A. bisporus vs. *P. florida*	1	345.9	345.9	10.0**
replicates	2	6.5	3.2	0.1
runs	2	373.6	186.8	5.4**
samples&treatment (ST)	3	7653.2	2551.0	73.7**
replicates X runs	4	15.2	3.8	0.1
replicates X ST	6	103.8	17.3	0.5
runs X ST	6	626.1	104.4	3.0
replicates X runs X ST	12	187.5	15.6	0.4
error	35	1210.8	34.6	
total	71	10522.7	0	

* significant at 5% level

** significant at 1% level

11. APPENDIX B

This appendix contains the score sheets which were used for the taste panel evaluations of Pleurotus florida in the freezing, canning and drying experiments.

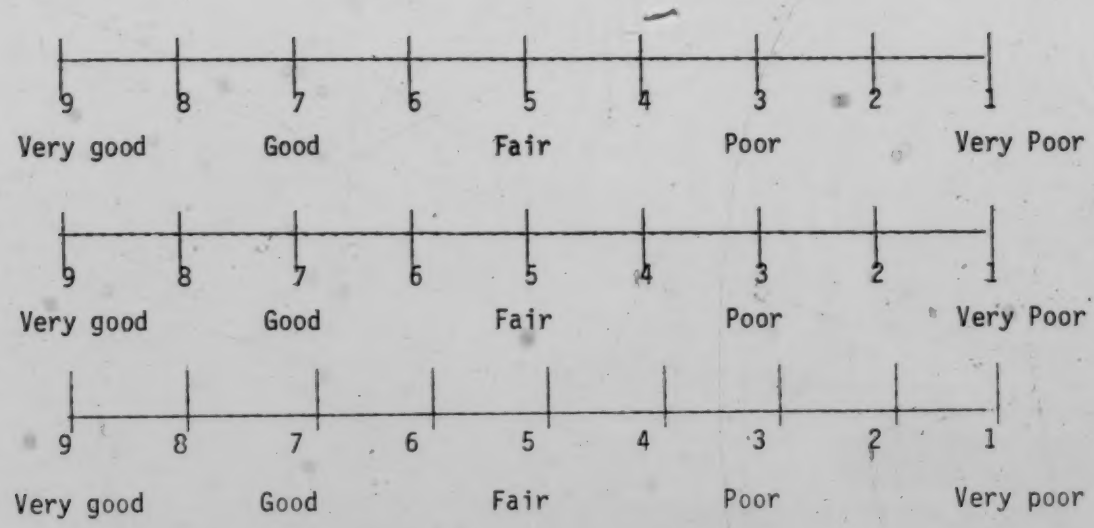
TASTE PANEL SCORE SHEET #1

NAME _____ DATE _____

Instructions:

Place a check mark on the scale below to indicate how you rate the flavor of each sample.

Sample #


9 8 7 6 5 4 3 2 1
Very good Good Fair Poor Very Poor
9 8 7 6 5 4 3 2 1
Very good Good Fair Poor Very Poor
9 8 7 6 5 4 3 2 1
Very good Good Fair Poor Very poor

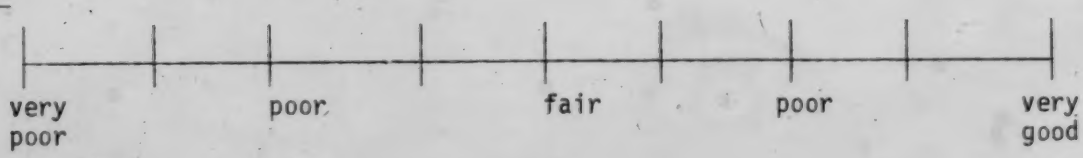
COMMENTS:

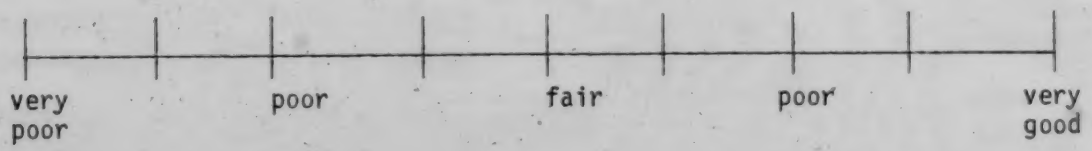
TASTE PANEL SCORE SHEET #2

NAME _____ DATE _____

Instructions: Place a check mark on the scale below to indicate how you rate the flavor of each sample.

Sample #





TASTE PANEL SCORE SHEET #3

NAME _____ DATE _____

Instructions:

1) Please indicate the degree of texture difference between the samples and the reference according to the following scale:

1 - considerably more tender
2 - moderately more tender
3 - slightly more tender
4 - no difference
5 - slightly less tender
6 - moderately less tender
7 - considerably less tender

Sample #	Score
_____	_____
_____	_____

2) Please indicate the degree of flavor difference between the samples and the reference according to the following scale:

1 - considerably more flavor
2 - moderately more flavor
3 - slightly more flavor
4 - no difference
5 - slightly less flavor
6 - moderately less flavor
7 - considerably less flavor

Sample #	Score
_____	_____
_____	_____

COMMENTS:

TASTE PANEL SCORE SHEET #4

NAME _____ DATE _____

Instructions:

Please rate the appearance, texture, flavor and overall quality of each sample according to the following scale:

1 - very poor
2 - poor
3 - fair
4 - good
5 - very good
6 - excellent

Please also add comments in the spaces provided to indicate characteristics you particularily liked or disliked in each sample.

Sample	
Appearance	
Texture	
Flavor	
Overall	
Comments	

TASTE PANEL SCORE SHEET #5

NAME _____ DATE _____

Instructions:

Please rate the appearance, texture, flavor and overall quality of each sample according to the following scale:

1 - very poor
2
3 - poor
4
5 - fair
6
7 - good
8
9 - very good

Please also add comments in the spaces provided to indicate characteristics you particularily liked or disliked in each sample.

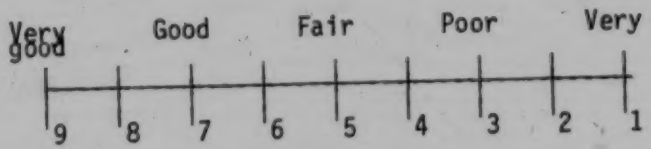
Sample #	
Appearance	
Texture	
Flavor	
Overall	
Comments	

TASTE PANEL SCORE SHEET #6

NAME _____ DATE _____

Instructions:

You will be given three samples to evaluate for flavor and consistency. Taste the samples and then score these attributes for each sample using the following scale:



Sample #	Flavor Score	Consistency Score

Comments:

_____

_____

_____

_____

TASTE PANEL SCORE SHEET #7

NAME _____ DATE _____

Instructions:

You are receiving two samples to compare for flavor and texture. You have also been given a reference sample, marked R, to which you are to compare each sample. Test each sample and score according to the appropriate scale. Then mark the amount of difference that exists.

Flavor

Sample #		
Better than R		
Equal to R		
Inferior to R		

Amount of Difference:

None	_____	_____
Slight	_____	_____
Moderate	_____	_____
Much	_____	_____
Extreme	_____	_____

Texture

Sample #		
More tender than R		
Equally tender		
Less tender than R		

Amount of Difference:

None	_____	_____
Slight	_____	_____
Moderate	_____	_____
Much	_____	_____
Extreme	_____	_____

Which sample is most acceptable to you in terms of texture? _____

COMMENTS:

TASTE PANEL SCORE SHEET #8

NAME _____ DATE _____

Instructions:

Here are three samples for evaluation. Two of these samples are duplicates. Separate the odd sample for flavor difference only.

Sample Code #	(Check Odd Sample)
_____	_____
_____	_____
_____	_____